

## **NORMATIVE INSTRUCTION No.32, OF MAY 13<sup>TH</sup>, 2002**

Technical rules of surveillance for the Newcastle disease and avian influenza, and of control and eradication of the Newcastle disease

**THE SECRETARY OF ANIMAL AND PLANT HEALTH AND INSPECTION, OF THE MINISTRY OF AGRICULTURE, LIVESTOCK AND FOOD SUPPLY**, by the powers invested in him in article 83, clause IV, of the Internal Regulation of the Secretariat, approved by Ministerial Ordinance No.574, of December 8<sup>th</sup>, 1998, in consideration and in light of the provisions of the Ministerial Ordinance No.193, of September 19<sup>th</sup>, 1994, and in File MA 21000.006729/2001-59, decide:

Article I To approve the Technical Rules of Inspection for the Newcastle disease and Avian Influenza, and of control and eradication for the Newcastle disease.

Article II This Normative Instruction will be in effect on the date of its publication.

Article III Is hereby repealed the SDA Ordinance No.183, of November 8<sup>th</sup>, 1994.

LUIZ CARLOS OLIVEIRA

PUBLISHED IN THE DOU No.91, SECTION 1, OF MAY 14<sup>TH</sup>, 2002

### **ATTACHMENT**

#### **TECHNICAL RULES OF SURVEILANCE FOR THE NEWCASTLE DISEASE AND AVIAN INFLUENZA, AND OF CONTROL AND ERADICATION OF THE NEWCASTLE DISEASE.**

##### **Chapter I Introduction**

1. The current rule defines surveillance measures for the Newcastle disease and avian influenza, and of control and eradication being applied to the Newcastle disease:

1.1. In the surveillance at the port of entry, focusing on the health control of poultry and generic material from imported poultry.

1.2. At aviary establishments permanently controlled, and eventually controlled.

1.3. At properties that maintain poultry for commercialization or for raising, at backyard birds farms and other poultry lodging locations for the ones kept in captivity.

##### **Chapter II**

## Diseases

1. THE NEWCASTLE DISEASE: is an infectious poultry disease caused by a virus from the Paramixovirus family, aviary Rubulavirus type of serotype 1 (APMV1), representing one of the following criterions of virulence:

- the virus has a pathogenicity index inside the brain of at least 0.7 on one day old chicks (*Gallus gallus*); or
- the presence of multiple basic amino acids is shown in the virus (directly or by deduction), in the C-terminal fraction of the F2 protein, or the same as the presence of phenylalanine in the 117 residue, which is the N-terminal fraction of the F1 protein. The term “multiple basic amino acids”, refers to at least three arginine or lysine residue, between residues 113 and 116.

In this definition, the amino acid residues are numbered from the N-terminal fraction of the amino acid sequence deducted from the nucleotide sequence of the gene. F0 and residues 113-116, correspondents to residues – 4 to –1, from the cleavage area.

In case the typical aminoacid residues cannot be distinguished, such as the description above, it is suitable to distinguish the isolated virus determining the pathogenicity index in the brain (RESOLUTION No.XIII, of May 1999, issued by the international OIE committee; OIE Zoosanitary Code, 2001).

2. AVIAN INFLUENZA: is a poultry infectious disease that is caused by a virus of the Orthomixoviridae family, of Influenzavirus A, B that present an Intravenous Pathogenicity Index (IPIV) > 1.2 on chicken that are 6 weeks old; or an infection caused by an Influenza A virus of subtype H5 or H7, with a sequence of nucleotides that present multiple basis of aminoacids in the cleavage area of the hemoagglutinin ((Manual Standards of Diagnostics Test and Vaccines OIE, chapter 2.1.14 year 1996; OIE International Zoosanitary Code, 2001).

## Chapter III Definitions

1. For the purpose of these rules, it is understood:

1.1.OIE: International Office of Murrains;

1.2.MAPA: Ministry of Agriculture, Livestock and Food Supply;

1.3.SDA: Secretariat of Animal and Plant Health and Inspection;

1.4. DDA: Animal Defense Department;

1.5. CLA: Animal Laboratory Coordinating Unit;

1.6. CPS: Surveillance and Health Program Coordination;

1.7. PNSA: National Plan of Poultry Health;

1.8. DIPOA: Animal Origin Products Inspection Department;

1.9. DFA: Federal Agricultural Delegacy;

1.10.SSA: Animal Health Service;

1.11.SIF: Federal Inspection Service;

1.12.OFFICIAL SERVICE: is the federal, state, and municipal animal health protection service;

- 1.13.OFFICIAL LABORATORIES: are the laboratories from the MAPA chain;
- 1.14.CERTIFIED LABORATORIES: are the laboratories of other federal, state, municipal, or private institutions, that have been approved and recognized by the MAPA, to perform laboratory diagnosis of disease agents referred by these rules;
- 1.15. FEDERAL INSPECTOR: is a MAPA inspector with veterinary qualifications that inspects and supervises matters regarding animal health protection;
- 1.16.OFFICIAL VETERINARIAN: is the Federal Inspector with veterinary qualifications or the official veterinarian of the official animal health protection service;
- 1.17.CERTIFIED VETERINARIAN: is the official, state, municipal, private, or an independent professional veterinarian, whom received delegation of competence of the federal official service to issue the Animal Transit Guide (GTA) or similar;
- 1.18.TECHNICIAN IN CHARGE: is the veterinarian responsible for the hygienic/health control at the flocks of the poultry farming premises, properly registered at the MAPA;
- 1.19. HEALTH CERTIFICATES: are health inspection certificates;
- 1.20.ANIMAL TRANSIT GUIDE (GTA): is the mandatory document used for the transit of poultry, hatching eggs and 1 (one) day old poultry, any type of handling and objective;
- 1.21.FORM IN: is the standard DDA form, used for opening the outbreaks and to perform an epidemiological investigation;
- 1.22.FORM COM: is the standard DDA investigation supplement form;
- 1.23.STATES OF THE PROGRAM: are the states named by the DDA, that develop health monitoring operations for the PNSA diseases and the permanent epidemiological surveillance for aviary diseases;
- 1.24.EPIDEMIOLOGICAL UNIT: it is a unit of the aviary establishment that allows the poultry that are lodged there to be treated and fed separately by distinctive personnel and other employees;
- 1.25. MONITORING OF FLOCKS: is the laboratory health and analysis follow up, by means of serological tests and other by other means, on other biological material or not, and epidemiological analysis of the health conditions of poultry lodged in an aviary establishment and adequate interpretation of the results;
- 1.26.COMMERCIAL BIRDS: generation of poultry used for producing meats, eggs, derivatives and subproducts;
- 1.27.SUSCEPTIBLE POULTRY: involves all domestic, wild, exotic and ornamental birds;
- 1.28.INFECTED POULTRY: is any poultry which has been officially tested positive for the Newcastle disease or avian influenza through conclusive laboratory proof;
- 1.29. LIVESTOCK PRODUCT: includes meat, eggs, legs, blood, bowels and bones from the susceptible animal;
- 1.30.POULTRY MEAT: it is poultry meat, muscular eatable part of the slaughtered birds, declared proper for human consumption by an official veterinary inspection performed before and after the slaughter;
- 1.31.CARCASS: is the full body of a poultry after numbness or not, bleeding, plucking and evisceration, in which crop, trachea, esophagus, intestines, cloacal, spleen, reproductive organs and lungs have been removed. The extraction of kidneys, feet, neck and head is optional;
- 1.32.SUBPRODUCTS: meat, blood, leg and bowel flours; incubation residues; aviary bed; skin; feather and plume; and phaners;
- 1.33.VEHICLE: any type of transportation on ground, water or air;

- 1.34.OUTBREAK: is the establishment in which the presence of the Newcastle disease or avian influenza has affected one or more birds;
- 1.35.PERIFOCAL AREA: is the area around the outbreak, which limits are established by the official service;
- 1.36.PROTECTION ZONE: ie the area of 3 (three) km radius around the outbreak, considered as the infected zone;
- 1.37.SURVEILLANCE ZONE: is the area of 7 (seven) km radius from the protection zone around the outbreak;
- 1.38. PROTECTION ZONE + SURVEILLANCE ZONE: 10 (ten) km radius around the outbreak;
- 1.39. DEPOPULATION: is the period of which the lodging of an aviary establishment is emptied out after the occurrence of an infestation, eliminating all birds and washing and disinfecting the barn;
- 1.40. HEALTH SLAUGHTER: is the slaughter of all diseased birds with signs of contamination or related to the matter of biosafety, its direct and indirect contacts;
- 1.41. EXTINGUISHING: elimination of poultry, its products, subproducts, meat or carcass, by any physical or chemical means that assures total inactivation of the Newcastle and avian influenza viruses;
- 1.42.EMERGENCY VACCINATION: is the vaccination applied as means of controlling the disease after one or more cases have been registered, or when the epidemiological or health situation require;
- 1.43.PROPRIETOR: all those who are depositary, or by any means , maintains in their possession one or more susceptible birds;
- 1.44.PROPERTY: location where birds are lodged for commercialization or not (e.g.: establishment where birds are lodged, for leisure or domestic breeding, and commercial stores);
- 1.45.AVIARY PREMISES: is the location where poultry is kept for any purpose, which may be constituted of one or more centers.
- 1.46.AVIARY ESTABLISHMENT PERMANENTLY CONTROLLED: they are the farms, genetically selected, of primary breeders (pure breeds), great grandparent farms, grandparent farms, head office farms, farms of breeding poultry free of specific pathogens (SPF), and the incubators of these establishments.
- 1.47.AVIARY ESTABLISHMENT EVENTUALLY CONTROLLED: they are the aviary establishments producing commercial eggs, poultry for slaughter, exploring other wild birds, or ornamental, and/or exotic or not, and the incubators of these establishments.
- 1.48.BARN: is the aviary production unit, characterized as the unit of a center, which lodges a group of breeders, poultry for slaughter or commercial laying hens, of the same age (with exception of pure breeds, genetically selected), and of the same breed.
- 1.49.CENTER: is the unit with an area adequately isolated, of common handling, constituted of one or more barns.
- 1.50.BATCH: group of poultry for the same purpose, origin and age, lodged in one or many barns.
- 1.51.BOXES: are physical sections divided inside a barn.

Chapter IV  
Procedures that must be complied by aviary premises

1. To attend to the PNSA, the aviary establishments of permanent and eventual controls must:

1.1. Be registered at the DFA, or registered in the cases established by the MAPA in the official service of the state in which they are located;

1.2. Be under the surveillance and control of the SSA/DFA, or of the State Secretariat of Agriculture, or the department that issued this document, in the state which they are located;

1.3. In the cases defined by the MAPA legislation, they must be supervised by a veterinarian responsible technician, registered at the DFA, or of State Secretariat of Agriculture, or by the department that issued this document, in the state which they are located, when the responsibility is passed over;

1.4. Proceed with the notification to the health authorities of any probable signs of the Newcastle or avian influenza diseases;

1.5. Use only immunogens, disinfectants, antigens, control serums, and “kits” registered at the MAPA, observing the starting number, name of the manufacturer, and expiration date.

## Chapter V Notification

1. The veterinarians, proprietor, or any other person that have knowledge of the occurrence or probable signs as to the cases of the Newcastle disease and avian influenza, are enforced to communicate the occurrence immediately to the official service (Decree No.24.548, of July 3<sup>rd</sup>, 1934, and Ministerial Ordinance No.070/94, of March 3<sup>rd</sup>, 1994).

1.1. The notification may be done personally, by telephone, radio, fax, e-mail or any other available way.

2. The infraction as per item 1 will be investigated by the official service that will use available measure to investigate responsibilities.

2.1. In the case of a veterinarian, besides what is mentioned or as per item 2, the official service must proceed according to the specific professional legislation.

The official service must be notified of the probable signs, preferably through the local veterinarian service, and sent to the an official laboratory or to one certified by the MAPA, for this matter, from any wound material caused by the disease that should be found during the inspection, the slaughtering or during the necropsy.

4. At the slaughterhouses, in cases of noticing signs of the disease(s), the slaughtering must be suspended until the conclusion of the cleanup and disinfection as recommended according to the criterions established by the DIPOA, and immediately communicated to the official service.

## Chapter VI Operation strategies

1. The monitoring of the Newcastle disease and avian influenza, and the control and eradication of the Newcastle diseases will be applied in all states of the country.

1.1. Since the avian influenza is considered an exotic disease at industrial poultry flocks in Brazil, the risk of entry must be evaluated in the country, and permanent surveillance must be kept.

1.2. Because of the economical importance of the aviculture and of the epidemiological characteristics, a study on viral activity will be performed with the focus of applying free zone from the Newcastle disease in the area of industrial production in the country, and the surveillance for the Newcastle disease and avian influenza in the states considered priority by the PNSA/DDA/SDA/MAPA.

2. The prophylaxis, the control and the eradication of these diseases consist in the application of the following measures of animal health protection:

2.1. Notification of suspicious signs of the Newcastle disease and avian influenza;

2.2. Assistance to the outbreaks;

2.3. Adopting biosafety measures;

2.4. Applying measures of disinfection;

2.5. Health sacrifice;

2.6. Depopulation;

2.7. Epidemiological analysis;

2.8. Routine or emergency vaccination at flocks;

2.9. Control and inspection of susceptible animals;

2.10. Transit control;

2.11. Other health measures.

## Chapter VII

### Assistance to the outbreaks

#### 1. PROBABLE SIGNS:

1.1. All probable signs and occurrence notifications for the Newcastle disease and avian influenza is secondly considered exotic at industrial poultry flocks of the country, must be immediately investigated by the official service following the rules of health protection, sending the samples to the an official laboratory, or to one certified by the MAPA, for this matter.

1.2. The notification of the probable signs of these diseases will include adopting the following health measures:

1.2.1. Interdiction of the aviary property or premises, opening of the FORM IN, and adoption of specific health measures with immediate collection of samples to be sent to a laboratory of the official chain or when authorized by the MAPA, for laboratory certified by the MAPA, for this matter, along with a copy of the FORM IN;

1.2.2. Registration of all categories of poultry indicating the number of dead birds, with and without clinical sign(s) of the disease(s) by category;

1.2.3. Poultry maintenance at lodging, or other confinement nuclei established by the federal farming inspector or by the official veterinarian, where they can remain isolated and the handling prohibited;

1.2.4. Control by the federal inspector or by the official veterinarian, of any transit of people, animals, vehicles, meats, carcasses, debris, excrement, beds, fomites and structures that might propagate the disease(s);

1.2.5. Utilization of adequate means to disinfect roads and exits of each aviary lodging premises, according to the recommendations of the OIE;

1.2.6. Conducting the epidemiological investigation with the opening of FORM IN, and later of FORM COM, to determine the origin of the infection and its propagation;

1.2.7. Abduction of the poultry meat and the eggs produced in the period of incubation of the diseases.

## 2. CONFIRMATION:

2.1. With the confirmation from the laboratory diagnosis of the Newcastle or avian influenza diseases, defined in Chapter II, of this rule, by means of conclusive laboratory exams, at the property where the outbreak was identified will adopt the following measures by the federal inspector or by the official veterinarian:

2.1.1. Immediate slaughter at the location of all the poultry from the aviary premises;

2.1.2. Extinguishing of all poultry that have died or have been sacrificed;

2.1.3. Extinguishing or appropriate treatment of all residues, such as: ration, bed and feces, and susceptible fomites of being contaminated;

2.1.3.1. The treatment must be conducted in agreement with the instructions of the federal inspector or by the official veterinarian, in a way to assure the extinguishing of the viruses of the Newcastle disease and avian influenza.

2.1.4. Extinguishing of the meat of all poultry originated from the farm and slaughtered during the period of the incubation of the disease;

2.1.5. Extinguishing of the eggs and sub products that were produced during the probable period of disease incubation;

2.1.6. Cleanup and complete disinfection of the breeding nuclei;

2.1.7. Establish the depopulation for at least 21 (twenty one) days before reintroducing poultry into the aviary establishment, initiated after the disinfection process is complete;

2.1.8. The measures for the epidemiological evaluation and health risk may be applied at other aviary establishments if instructed to do so by the official service;

2.1.9. The official service will proceed with the epidemiological investigation at all properties with poultry, aviary establishments and other lodging locations, protection zone, constituted in a 3 (three) km radius and a 7 (seven) km radius around the outbreak, from the protection zone (surveillance zone), determined with basis on fact of geographical, administrative, ecological and epizootiological origin related to the disease, registering all visits and stated occurrences;

2.1.10. The official service will establish the prohibition of transit and taking the poultry off the aviary properties, within the surveillance zone, for the minimum period of 21 (twenty one) days, except for those for slaughter sanitation, preferably at slaughterhouses with the SIF, located within the surveillance zone, designated and supervised by the federal inspector or official veterinarian.

3. The location in which the abducted material is located will be cleaned and disinfected, and products and sub products will be extinguished.

## 4. PROTECTION ZONE:

4.1. Measures must be adopted regarding the protection zone, as follows:

4.1.1. Immediate visit by the official service and future monitoring of all properties with poultry, aviary establishments and poultry lodging locations, performing clinical evaluation on the lodged birds, and collection of samples for laboratory examinations, registering all visits and occurrences;

4.1.2. Maintenance of all poultry in there lodge or any other location that allows isolation, at the choice of the official service;

4.1.3. Utilization of appropriate disinfection systems according to the criterions of the official service at port of entries and exits of the property or aviary establishment;

4.1.4. The official service will proceed with the transit control within this zone, of people, of material, equipment and vehicles that represent a health risk;

4.1.5. The official service will prohibit the transit and removal of poultry, eggs, manure, ration, sub products of poultry, fomites of the property or of the aviary establishment in which they are found, except with the authorization of the official service in charge of transportation, in the following conditions:

4.1.5.1. Poultry for slaughter is preferably at a slaughterhouse with the SIF, located at the infected area or, if that is not possible it must be performed at an evaluated location, designated and supervised by the federal inspector or by the official veterinarian;

4.1.5.2. One day old chicks or birds for an exploration located inside the surveillance zone and which does not contain other lodged birds;

4.1.5.3. The eggs for incubation and for hatching in an incubator within the protection or surveillance zones previously designated by the federal inspector or by the official veterinarian, controlled and performed in separate machines.

4.1.5.3.1. The eggs and their packages must be disinfected before transferring them to the incubatory.

4.1.6. The previously mentioned transfers must be performed directly by the official service and authorized after the health inspection on the property or aviary establishment, performed by a federal inspector or by an official veterinarian;

4.1.7. The means of transportation being applied must be cleaned and disinfected before and after its usage;

4.1.8. The poultry manure withdrawn from the FEED and the poultry sub products is subject to the control by the official service as to transportation and destination, after substantial evaluation, and is no longer represents a dissemination risk of the disease(s);

4.1.9. The official service will prohibit fairs, markets, exhibitions and any other poultry gatherings;

4.1.10. The official service will proceed with the introduction of sentinel poultry onto the depopulated outbreak property;

4.1.11. The official service will establish the serological control at official or MAPA certified laboratories for this matter, every seven days on sentinel poultry until the period of depopulation is complete, with the minimum of 21 (twenty one) days;

4.1.12. The measures applied in the zone of protection will be kept until the laboratory diagnosis is concluded, and the epidemiological investigation, for at least 21 (twenty one) days after the result, at the infected property or aviary establishment, of the preliminary cleaning and disinfecting operations, or by determination of the official service. After these measures, the protection zone will then be part of the surveillance zone.

## 5.SURVEILLANCE ZONE:

5.1.Measures must be adopted regarding the protection zone, as follows:



- 5.1.1. Investigation on all properties with poultry, aviary establishments and poultry lodging locations, in a 10 (ten) km radius, registering all visits and occurrences;
- 5.1.2. Prohibition by the official service of poultry and egg maneuvering within the zone, in the first 15 (fifteen) days;
- 5.1.3. Maintenance of all poultry in there lodges or any other location that allows isolation, at the choice of the official service;
- 5.1.4. Prohibition by the official service of the maneuvering and removal of poultry from the property or aviary establishment within the surveillance zone, except the ones intended for slaughter sanitation, preferably at a slaughterhouse with SIF, located within the surveillance zone or nearby, analyzed and designated by the federal inspector or by the official veterinarian;
- 5.1.5. Prohibition by the official service to remove eggs from the surveillance zone, saved if sent to an incubatory for incubation and hatching, evaluated and designated by the federal inspector or by the official veterinarian, with controlled incubation and performed in separate machines;
  - 5.1.5.1. The eggs and their packages must be disinfected before transferring them to the incubatory.
- 5.1.6. Prohibition for removing and using the manure, FEED and poultry sub products without an authorization of the official service;
- 5.1.7. The official service will prohibit fairs, markets, exhibitions and any other poultry gatherings;
- 5.1.8. The official service will proceed with the transit control within this zone, of people, of material, equipment and vehicles that represent a health risk;
- 5.2. The measure applied to the surveillance zone will be maintained until the conclusion of the laboratory diagnosis and of the epidemiological investigation for at least 30 (thirty) days, determined by the official service after exploring the infected environment, preliminary cleaning and disinfecting operations.
- 6. The operations described in this chapter may circumscribe the areas of the establishment which create an epidemiological unit; as long as the official service guarantees that the disease(s) will not spread to the other non infected units.

## Chapter VIII

### Collecting Samples and Forwarding them for Laboratory Testing

- 1. Location and events where the collection of the material is performed:
  - 1.1. Of poultry originated from any country, at the port of entry (port, airport or border), in quarantine, at the time of the veterinary inspection for the entry discharge;
  - 1.2. In the probable signs of infection occurrences by the official service;
  - 1.3. Performing the study project of viral activity, with focus on applying the free zone for the Newcastle disease within the area of industry production in the country, and surveillance for the Newcastle disease and avian influenza, for the monitoring of domestic flocks by the official service of animal health protection, and inspection of livestock products, within the areas of competence.
- 2. Samples

2.1. For isolation and identification of the virus, samples must be collected from live poultry after the necropsy of the sacrificed poultry, or of the ones which died with clinical symptoms suggesting the Newcastle disease and avian influenza.

2.1.1. Live poultry:

2.1.1.1. Serum;

2.1.1.2. Cloacal swab:

2.1.1.2. Tracheal swab:

2.1.1.4. Fresh feces.

2.1.2. Necropsy poultry (collect aseptically, isolated or in “pool”):

2.1.2.1. Spleen;

2.1.2.2. Brain;

2.1.2.3. Heart;

2.1.2.4. Feces;

2.1.2.5. Liver;

2.1.2.6. Aqueous humour;

2.1.2.7. Bowel;

2.1.2.8. Proventriculus;

2.1.2.9. Lung / trachea;

2.1.2.10. Air sacs;

2.1.2.11. Oronasal swab;

2.1.2.12. Cecal tonsils.

2.2. In case of probable signs as to a outbreak, focusing on reducing the risk of dissemination and diffusion of the virus(es) of the disease(s) during the transportation to the laboratory, it is recommended to perform the necropsy at the location, with collection of the material and adequate storage to be sent to an official laboratory, or to a laboratory designated by the MAPA.

3. Collection and storage of samples

3.1. They must be collected in PBS, pH 7.2, containing antibiotics in the concentrations of:

ANTIBIOTICS (p/ml of PBS)	SAMPLE			
	SWABS		FECES	ORGANS
	Trachea	Cloacal		

<b>Penicillin</b>	<b>2000 UI</b>	<b>10000 UI</b>	<b>10000 UI</b>	<b>2000 UI</b>
<b>Etreptomycin</b>	<b>2 mg</b>	<b>10 mg</b>	<b>10 mg</b>	<b>2 mg</b>
<b>Gentamicin</b>	<b>50 µg</b>	<b>250 µg</b>	<b>250 µg</b>	<b>50 µg</b>
<b>Fungizone</b>	<b>1000 UI</b>	<b>5000 UI</b>	<b>5000 UI</b>	<b>1000 UI</b>

- 3.2. Properly identified, refrigerated, sealed and stored in isothermal boxes;
- 3.3. Forwarded with FORM IN or a standard DDA collection form, properly filled out;
- 3.4. They will be registered at official or MAPA certified laboratories for this matter, on a proper book, according to the model indicated by the CLA/DDA/SDA/MAPA;
- 3.5. When being sent to the serology, they must be refrigerated or preferably frozen. Blood samples with signs of clots will not be accepted;
- 3.6. When received, they must be divided in 2 (two) pieces and identified, one as proof and the other as counterproof;
- 3.7. The identification tag of the counterproof, according to the model indicated by the CLA/DDA/SDA/MAPA, will be fill out and sealed along with the samples for the counterproof; the seal will be of plastic material, numbered and secured.
- 3.8. At the end of the study project of the viral activity of the Newcastle disease, surveillance of the Newcastle disease and avian influenza, do not apply to items 3.6 and 3.7.
4. Conservation and stocks of goods
  - 4.1. The samples that are intended for the virological exams must be kept under refrigeration, preferably frozen until its processing.
  - 4.2. The samples intended for the serology must be kept frozen at  $-20^{\circ}\text{C}$ , until its processing.
  - 4.3. After the result has been issued, the samples must be kept frozen at  $-20^{\circ}\text{C}$ , for at least 30 (thirty) days.

## Chapter IX

### Laboratory diagnosis

1. The laboratory procedures and exams for the diagnosis of the Newcastle and aviary influenza diseases, are determined by strict rules of the SDA/MAPA, with the possibility of performing the following exams:
  - 1.1. Imonoenzimatic test (ELISA);
  - 1.2. Hemagglutination test (HA);
  - 1.2. Hemagglutination inhibition teste (HI);
  - 1.3. Estimated time for the death of the embryo (TMM);
  - 1.4. Intracerebral pathogenicity index (IPIC);
  - 1.5. Intravenous pathogenicity index (IPIV);
  - 1.6. Immunodiffusion on agar gel (AGP);

- 1.7. Molecular biology techniques.
2. Other exams will only be considered when approved by the PNSA/CPS/DDA/SDA.
3. Only laboratory exams related to the diagnosis of the diseases standardized by the MAPA will be accepted, performed at official or MAPA certified laboratories for this matter, and confirmed by the Laboratory of National Reference.
4. All professionals and laboratories performing diagnosis on aviary diseases are obligated to immediately notify the probable signs or occurrences of the Newcastle and avian influenza diseases.
5. All the material for laboratory examinations must be sent along with the FORM IN or by a standard DDA/SDA/MAPA collection form, fill out correctly, signed by the federal inspector or by the official veterinarian, or even by the person responsible for the collection endorsed by the official service.
  - 5.1. At the port of entry, the standard MAPA material importation form will be used.
  - 5.2. During the study project of viral activity of the Newcastle disease, and the surveillance of the Newcastle disease and avian influenza, a standard DDA/SDA/MAPA collection form will be used.
  - 5.3. In the case of probable signs as to an outbreak, the FORM IN will be used.

## Chapter X

### The forwarding of laboratory results

1. The laboratory test results must be issued on a standard MAPA form, and communications according to the flowchart established:
  - 1.1. Negative result: send FAX or other type of immediate notification to the DDA/SDA/MAPA, and to the SSA/DFA/MAPA of the state in which the establishment is located;
  - 1.2. Positive result: send FAX or other type of immediate notification to the DDA/SDA/MAPA, which will notify the SSA/DFA/MAPA.

## Chapter XI

### The study for viral activity of the Newcastle disease and surveillance for the Newcastle disease and avian influenza

Prophylactic activities will be performed, with focus on controlling entries of possible exotic disease agents into domestic territory such as avian influenza, during the permanent epidemiological and health surveillance of the Newcastle disease:

- 1.1. During the unloading process at the port of entry, during the health inspection of the genetic material (of poultry or hatching eggs), by the airport surveillance service (SVA/DFA/MAPA);
- 1.2. During the official quarantine or the incubation of the hatching eggs, by the official service.
2. The material collected from the one day old poultry, hatching eggs or cloacal and tracheal swabs from any country, will be forwarded to an official laboratory in a package sealed by the MAPA, to perform the laboratory examinations and identify the disease agents, along with the standard collection form.

3. The study project of viral activity for the Newcastle disease, and the surveillance for the Newcastle disease and avian influenza, for the monitoring of the domestic avian flocks throughout different states will be introduced by the DDA/SDA/MAPA, observing the status of epidemiological diseases, considering the exotic disease status for avian influenza at Brazilian flocks, poultry industries:

3.1. Initially it will be applied in the production area, with the possibility of being applied to other production systems according to the project evaluation, and by the determination of the DDA/SDA/MAPA.

3.2. The states participating in the project will be defined by the DDA/SDA/MAPA.

3.3. The periodic collections of blood serum, tracheal and cloacal swabs from the poultry of the same batch at slaughterhouses with SIF, with the possibility of collections at breeding establishments determined by the PNSA/CPS/DDA/SDA/MAPA, according to this project.

3.4. The laboratory exams provided will be serological, isolation and viral characterization tests;

3.5. The activities regarding the samples collected may be performed by the SSA, SIF of the DFA/MAPA, or by the State Departments of Agriculture, or by the ones responsible for these activities, when this activity is authorized in agreement with this project.

3.6. The serological exams in this project will be defined in the scope of the DDA/SDA/MAPA, as per the correlation between them.

3.7. The collections for monitoring and for diagnosis, will only be accepted when performed by the federal inspector or by an official veterinarian, or under their supervision.

3.8. The sampling, period of collection, serological tests, the analyses criterions related to vaccinated and non-vaccinated poultry and the interpretation of the results will be defined in the referred project.

4. The laboratory exams will be performed by the MAPA official laboratory, of national reference to these diseases, and may be performed at federal or state institution laboratories, or when authorized by the CLA/DDA/SDA/MAPA.

5. The epidemiological analysis will be performed according to the information established by the DDA/SDA/MAPA.

6. The result evaluations will be nationally performed at the DDA/SDA/MAPA.

## Chapter XII

### Cleaning and disinfecting measures

1. The cleaning and disinfecting measures used for the control of outbreaks, will proceed with the criterions established on the OIE manual, and in specific manuals of the PNSA/CPS/DDA/SDA/MAPA.

## Chapter XIII

### Vaccination

1. The systematic vaccination against the Newcastle disease is optional throughout the states, as per the local epidemiological status.
  2. According to the epidemiological status of each region, after the evaluation by the official service, the poultry vaccination against the Newcastle disease may be enforced on properties and permanently or eventually controlled aviary establishments, or also performed regularly.
  3. It is responsibility of the official service during emergency situations to establish vaccination schedules per area.
- The vaccination against these diseases will only be performed with registered and MAPA approved vaccines (Decree No.1.662, of October 6<sup>th</sup>, 1995, and Ministerial Ordinance No.186, of May 13<sup>th</sup>, 1997), as a measure of prophylactic order or control of the disease.
5. In the case of the avian influenza since it is considered an exotic disease in the country, the vaccination can only be performed when authorized by the DDA/SDA after confirmation of the disease, risk evaluation and analysis of the epidemiological status.

#### Chapter XIV Transit

With intensions of avoiding the introduction and the spreading of these diseases, at the time of issuing the GTA for susceptible poultry, or for the interstate transit of poultry for slaughter at slaughterhouses must be

demanded by the issuer, among others, the following conditions:

1.1. At production establishments of international market providers:

1.1.1. The birds must be from a property or aviary establishment which have not had any signs of infection of the Newcastle disease and avian influenza for the last 90 (ninety) days, and that the surroundings within a 10 (ten) km radius, has not been infected by these diseases in the past 30 (thirty) days;

1.1.2. After the epidemiological situation is noticed and Chapter XIII, of this rule, the requirements for the transit of susceptible poultry in locations considered of risk, must prove that they were not vaccinated against the Newcastle disease for at least 30 (thirty) days prior to the slaughter.

2. The poultry may be prevented from transit when unaccompanied of the GTA, issued as per these rules, will be prohibited for transit, set in accordance with these standards, the responsible authority must draw up the Term of Incident, and determine the return to the source, without prejudice to the appropriate sanctions.

3. To perform interstate transit, it is demanded that the GTA be used. For interstate transit, it is demanded that the GTA be used, being able to be used in exceptional cases that are justified, to the acceptance of the similar transit document established in the state scope.

4. The vehicles for the transportation of susceptible poultry must be washed and disinfected according to the orientation of the official service.

5. The transportation of poultry residues, sub products must be done in vehicles that are protected or closed.

#### Chapter XV Incubation control

1. Biosafety measures on the incubation, when determined by the official service:

1.1. The egg incubation must meet to carry out the provisions in Chapter VII, of this term, respecting the natural agreements in the control of the protection and surveillance zones;

It is forbidden to incubate hatching eggs from great grandparent, grandparent and parent hatchers, and in the same period, meeting all health criterions of the superior breed.

## Chapter XVI

### General dispositions

The SSA/DFA/MAPA of the State in which the aviary premises is located, and the State Departments of Agriculture, are the responsible and competent offices for defining appropriate measures for handling health issues, as per the established in the Animal Health Protection Regulation and in the PNSA.

2. In consideration to the characteristics of a health emergency in the occurrence of an outbreak of the Newcastle disease or avian influenza, and the need of the official service to apply measures of immediate eradication, the establishments that produce poultry free of specific pathogenics (SPF), must provide 10 birds used as sentinel upon official solicitation, which will be used as sentinels for evaluation and end of outbreak.

3. Since the avian influenza is considered an exotic disease in the domestic aviary industrial flock, they must be observed, investigated and evaluated at a laboratory, and epidemiologically by the official service, additionally as described in item 2, of Chapter II, in these rules, for the following situations:

3.1. Any influenza virus that is lethal to 6,7, and 8 susceptible 4-6 week old birds, 10 days after intravenous inoculation with 0,2 ml of cornea-allantoids liquid diluted at 1:10, bacteria free;

3.2. Any H5 or H7 influenza virus that does not meet the criterion of the previous item, however that has a sequence of amino acids (in the cleavage site of the hemagglutinin), that is compatible with the highly pathogenicity influenza virus;

3.3 Any influenza virus that is not H5 or H7, that kills 1 to 5 times (pathogenicity), and grows in a cell in the absence of trypsin.

3.4. After careful evaluation by the official service and by the PNSA/CPS/DDA/SDA/MAPA, health measures that are relevant to the case will be applied.

4. The omissive cases and the questions created in the application of this rule, and in complementary acts will be voided by the DDA/SDA/MAPA.