



FEDERAL REPUBLIC OF BRAZIL
Ministry of Agriculture, Livestock and Food Supply
Secretariat of Animal and Plant Health
Department of Animal Health

BRAZILIAN SYSTEM OF VETERINARY EMERGENCIES
SisBRAVET

ACTION PLAN FOR FMD

BRASIL

Programa Nacional de Erradicação e Prevenção da Febre Aftosa



VOLUME I

RESPONSE TO THE NOTIFICATION OF SUSPECTED VESICULAR DISEASE

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Opening Remarks

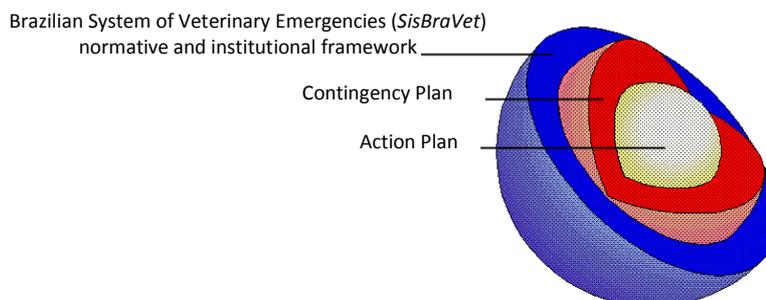
Emergency veterinary actions make up a complex set of activities involving an intricate network of technical, political, economical and social aspects. Therefore, implementing them entails planning and the definition of every aspect involved, in order to constitute a system of control and management. The constitution and maintenance of this system must consider various levels of organization and execution, adapted to the political and institutional reality of the country. In order to allow a simplified visualization of this system, at least three levels have been recognized :

Level 1: Normative and institutional framework of the system management. It includes the levels of higher hierarchy in the issues referred to the institutional relationship and to the activities of political, strategic, normative, regulating and coordinating nature, being able to include activities of operational nature if determined by national interest. The main purposes are to contribute to reach a high standard of good practices in managing veterinary emergencies in any point of the national territory; to ensure resources for development and maintenance of appropriate structures of surveillance and sanitary intervention; to guarantee the preparedness and maintenance of the available work force; and to maintain the continuous integration between the sectors and the institutions involved, in order to allow a rapid and effective response to animal diseases.

Level 2: Connection between the levels of top management and the field operational activities. It includes the consolidation and the organization of the guidelines and the general procedures of the system. It has the main purpose of ensuring the execution of the activities of veterinary surveillance, particularly the detection of infection sources and the coordination of the immediate response for control of the outbreaks and for reestablishment of the previous animal health status of the areas or regions affected. It also includes the level responsible for controlling and managing information and for defining and maintaining operational standards.

Level 3: Operational basis of the system made up by the set of surveillance activities and sanitary intervention appropriate and adapted to each type of disease. It has the purpose of implementing the policies and the sanitary guidelines defined and organized in the previous levels. It is also responsible for taking, registering and organizing information related to its jurisdiction.

Considering the main documents for regulation, organization and guidelines of the whole system, Level 1 is represented by the legal acts and by the institutional guidelines, Level 2 by the *Contingency Plan* and Level 3 by the *Action Plan*. In the following picture, adapted from the FAO Emergency Prevention System (EMPRES), a schematic vision of the levels in question are presented.



Representation of the three management levels of the Brazilian System of Veterinary Emergencies

The “Contingency Plan” can only be extended to the whole country if the “Brazilian System of Veterinary Emergencies” (SisBraVet) has organized resources, structures and procedures aimed at developing the capacity for immediate notification and immediate response of all levels of the Unified Animal Health System during a veterinary emergency.

This “Brazilian System of Veterinary Emergencies” must have an effective vertical integration between the Ministry of Agriculture, Livestock and Food Supply (MAPA) and the state veterinary services; and effective horizontal integration between the Secretariat of Animal and Plant Health of MAPA, the Secretariat of Civil Defence and the National Centre of Risk and Disaster Management linked to the Ministry of National Integration, since veterinary emergencies caused by highly contagious infectious agents are recognized as natural disasters and are incorporated in the *National Plan of Disasters*. This recognition is founded on the potential of the epidemic spread of the infectious agents with a capacity to produce serious sanitary, social and economical consequences, being able to affect national and international trade, food safety or public health. Therefore, MAPA is part of the National Council of Civil Defence (CONDEC) and participates in the National System of Civil Defence (SINDEC).

The “Action Plan” is the main operational tool of the National System of Veterinary Emergencies. It is made up by a set of instructions to be implemented during a veterinary emergency from the first alert (notification) of the suspect until its control or eradication. It must be adapted and drafted separately for each type of veterinary disease. The **Action Plan for FMD** is set forth in the present document.

Considering the various stages that involve the surveillance system for vesicular diseases, this Action Plan is presented in two volumes:

Volume I: “Response to the notification of suspected vesicular disease” and

Volume II: “Declaration and management of the veterinary emergency situation for FMD”

Volume I contains information and instructions for action in case of any suspect of vesicular disease. It includes the stages of “investigation” and “alert”, which every veterinarian in the official veterinary service must be familiar with and master.

Volume II gives specific instructions that must be adopted in case of confirmation of FMD. It comprises the “emergency” and “conclusion” stages, requiring specific training of the team of professionals that must be constantly ready to act in veterinary emergency actions.

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Action Plan for FMD

1. Introduction

As specified in the Opening Remarks, Action Plans are elements integrating the Brazilian System of Veterinary Emergencies (SisBraVet). They represent the main operational tool of this system, being made of a set of instructions to operate during a veterinary emergency, from the first notification of suspect until its eradication. In this case, the instructions refer to vesicular diseases particularly **FMD**.

Considering the constant improvements in the knowledge about how infectious diseases work and the control and eradication techniques available, Action Plans are constantly being revised. This version has been drafted based on the rules of the National Program for Foot and Mouth Disease Eradication and Prevention – PNEFA (Normative Instruction nº 44, of October 2nd, 2007); in the *Manual of Procedures for the Response to Foot and Mouth Disease and other Vesicular Diseases* (PANAFTOSA – PAHO/WHO, Series of Technical Manuals, nº 9, 2007); in the guides and technical guidelines produced by the World Organisation for Animal Health (OIE) and by the Food and Agriculture Organization of the United Nations (FAO); in the analysis of the emergency plans of various countries and in the accumulated experience in Brazil in emergency actions to eliminate or control FMD outbreaks (1998, 1999 and 2005/2006 in Mato Grosso do Sul; 2000 and 2001 in Rio Grande do Sul; 2004 in Pará and Amazonas) and 2005/2006 in Paraná). Its publication follows technical discussions with the participation of many departments of MAPA and state veterinary services.

Execution of Action Plans must be included in a context of institutional organization that entails political-strategic definitions, technical preparedness and availability of physical, human and financial resources.

Taking into consideration the geographic, livestock and social-economical diversity of the country, it is not possible to expect that the Action Plans completely meet every need during an emergency action. They are tools that help reveal the purposes and procedures in emergency situations. Due to diversities found in the field, there will always be a need for adaptation. In order to do this, professionals responsible for managing actions of veterinary emergency in the field must have the operational autonomy and the technical know-how required to make decisions.

The present Action Plan gathers the know-how and the basic procedures referred to the response to suspected vesicular disease and veterinary emergency actions to eliminate FMD outbreaks. Despite the diversity of conditions observed in the field, it must be the basis of all veterinary services in the country. **Mastering its content is compulsory for all experts involved and must be part of training on the surveillance system for vesicular disease.**

The plan is organized into two volumes, in accordance with the logical sequence of surveillance and detection of cases of the disease. This process is grouped in four stages, as described in Figure 1. *Investigation* and *Alert* must be known by all veterinarians in the official animal health system, and are described in Volume I of the Action Plan, while stages of *Emergency* and *Conclusion* are described in Volume II and requires specific training and is addressed to a specific and smaller group of professionals that must be part of the national group of veterinary emergency, with representatives of MAPA and of the state veterinary services. Each stage will be discussed and detailed below in this Action Plan. Additional detail for specific subjects are presented as appendices to this document.

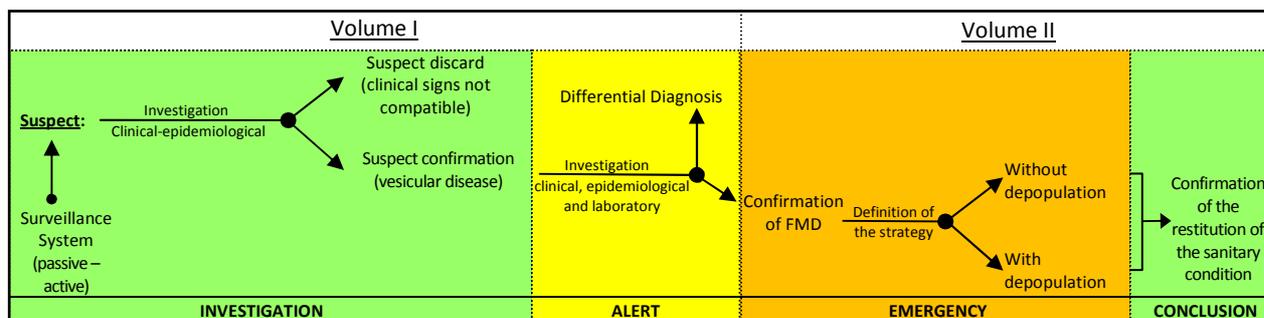


Figure 1. Representation of the main stages of the surveillance system for vesicular diseases.

Because the Action Plan is an integrating part of the surveillance system for FMD, it is indispensable that the experts involved know and master the *Veterinary Surveillance for vesicular diseases: general guidelines*, drafted by the Department of Animal Health – DSA/SDA/MAPA with a support of the state veterinary services. In addition, it is also necessary to constantly read technical and scientific texts on the subject, to master the legislation in force that rules the actions of animal health service and to know the guidelines and standards established by institutions that deal with this theme, such as the OIE (i.e.: Terrestrial Animal Health Code), the FMD Pan-American Centre - PANAFTOSA (i.e.: Manual of Procedures for the Response to Foot and Mouth Disease and other Vesicular Diseases; technical bulletins etc.) and FAO. All these institutions make an immense quantity of information available on the Internet. In PANAFTOSA website (www.panaftosa.org.br) there are links to the *Situation Room* and to the *Virtual Library*; on the OIE website (www.oie.int) there are links regarding the *Official Sanitary Status, Animals Diseases; Sanitary Rules; Technical disease cards* and *Worldwide Sanitary Information/Emergency Preparation*; and on FAO website (www.fao.org), the link for *System of Emergency Prevention of Diseases and Diseases of Animals and Plants (EMPRES)*.

In this first Volume, before beginning to describe the actions and activities related to each of the two initial stages (investigation and alert), in the item below will presented information and recommendations on the necessary structures, particularly to the state veterinary services.

2. Initial Recommendations

In order to improve the effectiveness of vesicular disease surveillance actions and the capacity of prompt response in emergencies to eliminate FMD outbreaks, in addition to a prepared staff, basic equipment and financial resources, there is a need to have information and specific structures as detailed below:

- a) Records of holdings and farmers are an indispensable piece of information. Every local veterinary unit must have an electronic file with the updated report of the holdings existing in their geographic jurisdiction, in accordance with the guidelines and the Standards of the Standardization Manual – CTQA/DSA, and in the Manual of Veterinary Surveillance of Vesicular Diseases – CFA/CGCD/DSA (available in MAPA's website: www.agricultura.gov.br). Every piece of information on herds susceptible to FMD must always be updated. Special attention must be given to the codification and geographical location system of the holdings (latitude and longitude), in accordance with the established standards. Every piece of information must help a great deal during the veterinary emergency actions;
- b) Database for GTA issuing are also an indispensable piece of information. The official veterinary service must prioritize the implementation of computerized systems to control holdings and farmers and to issue the GTAs (Animal Movement Permits). Computerizing Standards can be found in the *Standardization Manual - CTQA/DSA*.

Considering the information available on the holding records, issuing of GTAs and the peculiarities of each location, with technical and logistic support of the central coordination units of the state veterinary services, it is important for the professionals in charge of the local veterinary units to be familiar with the main production characteristics of the area under their jurisdiction, particularly with the entry and exit flows of animals, animal products and by-products. Information on these movements, including origin and destination, must be always updated for the whole state by the technicians of the central unit of state veterinary services. If an FMD outbreak is confirmed, all this characterization is very important to define the location and size of the area to be blocked and to investigate the activities at the place of origin of the infection;

- c) In the local veterinary units (considering their whole area of operation)¹:
 - i. List of names, positions, addresses and contact information of the municipal authorities (including the police), representatives of the civil defence and representatives of the agribusiness industry. When located in international or state borders, include names, addresses and contact information of the person in charge of neighbouring local veterinary units, belonging to the neighbour country or state;
 - ii. Arrangement of the animal disease emergency team in the state, with addresses and contact information of their representatives, particularly those responsible for bordering local veterinary unit of the neighbouring country or state;
 - iii. Contacts of the person in charge of municipal social communication and for the media available (TV, newspaper, radio), with names and addresses of every person in charge or representative;
 - iv. Records containing name, background, address and contact information of experts of private enterprise and other institutions acting in the field, chiefly veterinarians, animal science experts and agricultural technicians.
 - v. List of available heavy machines such as excavators, crawler tractors, power shovel, trucks with buckets, among others that can be used in the veterinary emergency activities (including names and how to contact the people responsible for releasing these machines);
 - vi. Hotel capacity and other places to lodge a large number of professionals;
 - vii. Available airports and landing strips, including those for small airplanes;
 - viii. List of places for possible implementation of the local unit to coordinate field activities during emergency actions (i.e. schools, gyms, community centers). The place has to be large and available to be used for at least three months in a row, and must have the following characteristics: capacity to hold a warehouse, some room that could be used as a parking place for many vehicles, good lighting and plenty of water, it must enable control of the entry of vehicles and people, a place for cleaning and disinfecting clothing and vehicles, rooms for working teams, a separate room for the coordinators and for holding technical meetings with telephone and internet;
 - ix. Identification of possible sites to place fixed inspection posts;
 - xi. Main access ways, including traffic conditions

¹ Information must be available in electronic format, when necessary, in order to allow a quick reference by the central unit of the state veterinary service.

- xii. Records of owners and drivers of vehicles that transport animals or risk products and of people responsible for transportation companies of animals in the region, including type, quantity and capacity of vehicles per owner or transportation company; and
- xiii. Specific records of settlements, indian tribes and “quilombola” communities;
- xiv. Location, including geographic coordinates, of establishments and points of interest to the animal health system, such as:
- Landfills, including management conditions;
 - Slaughterhouses, including slaughter capacity per species, and names, telephones and addresses of the veterinarians in charge;
 - Retailers of veterinary products, including the name of the veterinarians in charge;
 - Dairy and cooling plants (include the records of people in charge of milk lines and the route of each line);
 - Auction grounds and other animals gathering, with the identification of the owner and the responsible technician;
 - Rendering plants, tanneries and salting plants;
 - Farm hotels; and
 - Rural schools;
- b) Basic material for surveillance: every local veterinary unit must have appropriate transportation and communication media to operate in the area. They must also have response material in case of suspected vesicular disease and the list of material with the records of periodical checks carried out to ensure that everything is in good condition to be used any time. In order to do this, the person in charge of the local veterinary unit must be disciplined and the organized to systematically control the equipment available. If the material is incomplete, the professional has to officially notify his superior. Appendix 1 has a suggested form with a summary of the required material and instruments and with a field to record the control activities. A similar list must be kept in a visible place in every local veterinary unit. The control includes to check the expiry date of disinfectants, antiseptics and medium for sample preservation. In addition to the material summarized on Appendix 1, the state veterinary services must have material to collect oesophageal-pharyngeal fluid (preservation medium and Probang cups for adult and young animals) strategically distributed in the state.
- Equipment such as digital cameras and GPS are useful.
- c) Financial resources promptly available and procedures for rapid submittal of samples for laboratory testing: they are very important elements, and their lack may affect the whole work already made. Thus, the state veterinary services must have previously defined the shipping strategies of biological material from the field to the state central unit and from there to the National Animal and Plant Laboratory (LANAGRO) that will perform the laboratory testing. The possibility for material to be directly transported by the state veterinary service (personally) from the local unit to the central unit and from the central unit to LANAGRO should be considered. Irrespective of the path chosen, the material shipping must be preceded by telephone contact with the people in charge at LANAGRO, in order to agree on details of time and delivery, which must be confirmed by fax or e-mail. For this reason, each state veterinary service must keep an updated list of contact addresses (e-mail, telephone, and fax) of the official diagnostic network of MAPA.
- d) Geotechnologies: geographic data tools are very important to support veterinary emergency activities. The state veterinary services must invest in staff capacity building and in purchasing GIS (Geographic Information Systems) software, pinpointing that free software and a large basis of geographic data is currently available. Every local veterinary unit must have printed maps of their geographic jurisdiction in appropriate scales (1:50,000 or greater for better detail), including information on the geopolitical borders, roadways, waterways, location of holdings, villages, small towns, indian tribes, settlements, preservation or environmental protection areas, forest reserves, among other important elements for sanitary intervention activities. The field team for veterinary emergencies must count on the support of geosciences experts.

3. Stage 1: Investigation

3.1. Definition of Vesicular Disease

The definition of vesicular disease, among other diseases that are relevant for surveillance and disease emergency activities, was updated by Normative Instruction n° 44, of October 2nd, 2007, in accordance with the OIE international standards and is written down below:

- “Types of cases in the investigation of vesicular diseases:
 - a) *suspect case of vesicular disease*: notification made to the official veterinary service indicating the possibility of existing one or more animals with clinical signs compatible with infectious vesicular disease;
 - b) *probable case of vesicular disease*: the official veterinary service verified that there are animals with clinical signs of infectious vesicular disease, requiring immediate Biosecurity measures and preparation for laboratory diagnosis;
 - c) *case of vesicular disease ruled out*: every suspect case of vesicular disease investigated by the official veterinary service which clinical signs are not compatible with infectious vesicular disease;
 - d) *FMD case or outbreak*: recording, in an epidemiological unit, at least one case meeting at least one of the following criteria:
 - Isolation and identification of the FMD virus in samples from susceptible animals, with or without clinical signs of the disease, or in products from these animals;
 - Detection of the viral antigen specific of the FMD virus in samples coming from confirmed cases of vesicular disease, or from animals that could have had previous direct or indirect contact with the aetiological agent;
 - Existence of an epidemiological link with other FMD outbreak, finding at least one of the following conditions:
 - Presence of one or more probable cases of vesicular disease;
 - Detection of antibodies against structural (or capsidal) proteins of the FMD virus in animals not vaccinated against this disease; or
 - Detection of antibodies against non-structural proteins (or non-capsidal) of the FMD virus in animals, if the hypothesis of infection cannot be ruled out by epidemiological investigation;
 - e) *Case of FMD ruled out*: all probable case of vesicular disease that does not meet the criteria for confirmation of case or outbreak of FMD.”

The definition of case follows a coherent flow of investigation of the suspects of vesicular disease, in accordance with the graph in Figure 1 (page 2, Introduction). Another simplified graph, considering the types of cases defined for vesicular disease, is presented in Figure 2.

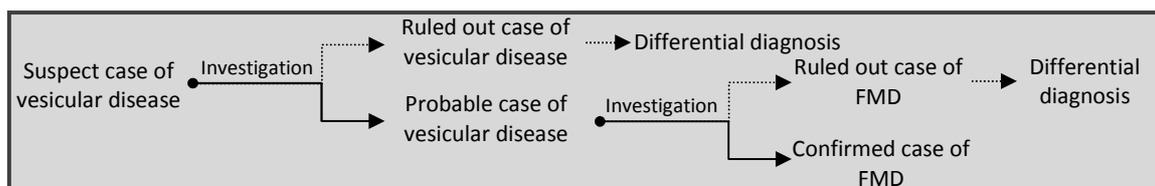


Figure 2. Representation of the investigation flow of suspect cases of vesicular disease

3.2. General considerations on the investigation of vesicular diseases

With regard to the passive surveillance system, the stage of investigation starts when a communication of suspect of vesicular disease is received by the official veterinary service.

Each suspect case of vesicular disease must be investigated by the official veterinary service within twelve hours (§3, art. 4, Normative Instruction nº 44 of October 2nd, 2007) irrespective of its origin. The result of the investigation must confirm or rule out vesicular disease. Among the ruled out cases, there are the ones of traumatic origin, intoxications and other infectious diseases that cannot be included in the definition of infectious vesicular disease. The probable cases of vesicular disease require supplementary investigation, including sampling of material for laboratory diagnosis, and designate the beginning of the alert stage that will be detailed in this Volume.

The clinical and epidemiological assessment of the suspects represents a decisive stage in the veterinary surveillance system of vesicular diseases. The veterinarian of the official service must be prepared to rule out or confirm the suspect of vesicular disease, requiring knowledge on the pathogen and the epidemiology of vesicular diseases, field experience and semiology techniques.

The table below describes the main stages of FMD pathogen and Figure 3 is a graphic representation of the theoretical evolution of the biological reactions expected in a non-vaccinated animal post-infection, highlighting the ideal moments for material sampling for virus isolation. This information has been adapted from materials prepared by PANAFTOSA.

FMD Pathogen	
a – Virus Inhalation b – Cells infection on the nasal cavity, pharynx and oesophagus c – Virus replication and spread to adjacent cells d – Passage of the virus to blood and lymph vessels e – Infection of lymph nodes and other glands f – Infection of cells in the oral cavity, feet, udder and rumen	24 - 72 h (1 to 3 days)
g – Onset of fever h – Appearance of vesicles on the oral cavity, feet, udder and rumen i – Drooling, nasal discharge and lameness	72 - 96 h (3 to 4 days)
j – Vesicles rupture and intensification of symptoms k – End of fever l – End of viremia and beginning of antibody production	120 h (5 days)
m – Reduction in the titer of virus in many tissues and fluids	as of 8th day
n – Cure of lesions and the animal starts to eat	as of 10th day
o – Gradual disappearance of the virus from tissues and fluids p – Increase in antibody production	as of 15th day
q – Complete cure (Virus may survive in the nasal-pharyngeal region for 6 to 24 months in cattle and 4 to 6 months in small ruminants, in accordance with the OIE technical disease cards)	15 days

Source: www.panaftosa.org.br (Links: Vesicular Diseases/FMD/Diagnoses, accessed in July, 2008)

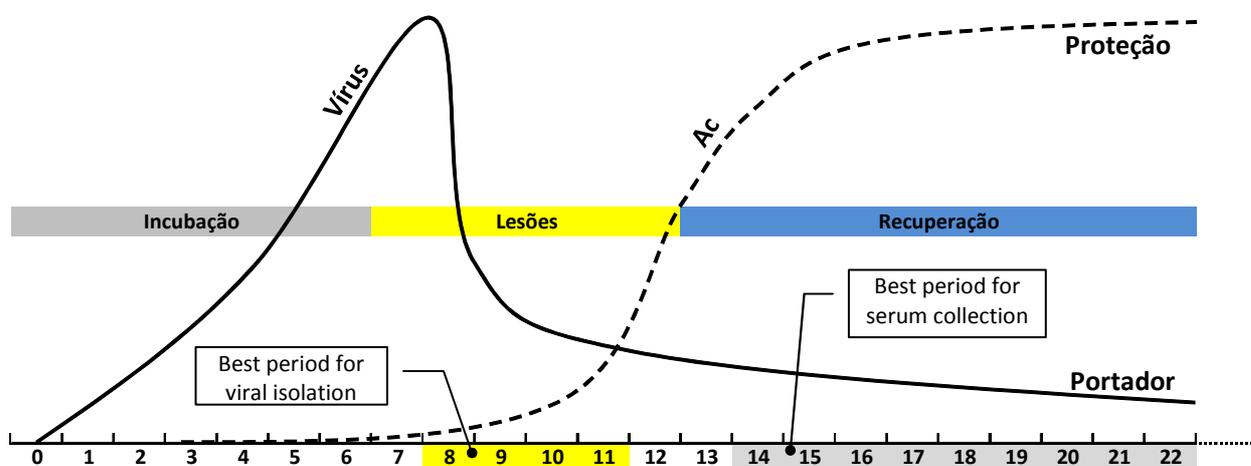


Figure 3. Theoretical evolution of FMD in infected cattle.
(Adapted from the document Series of Training Manuals, nº 2, PANAFTOSA, 1978)

Between the introduction of the virus (intracellular penetration) and the appearance of the first lesions, there is the incubation period ranging from 2 to 14 days, being characterized by two different stages: eclipse stage and prodromal stage. In the eclipse stage, the virus cannot be found even if sophisticated methods of investigation are used. This stage can last a few hours and corresponds to the intracellular penetration of the agent and formation of the first few virions². As soon as these virions are spread throughout the organism through blood (viremia) and lymphatic pathways, the prodromal stage starts and lasts until the appearance of the typical FMD lesions. In the prodromal stage, the animals show non-specific signs (febrile reaction, depression and anorexia), common to many infectious diseases.

Vesicular diseases with indistinguishable clinical signs of FMD are: vesicular stomatitis, swine vesicular disease and vesicular exanthema of swine. Appendices 2-6 have graphs and technical texts available on the OIE website with summarized information on the FMD, vesicular stomatitis and swine vesicular disease (note that they are summaries, with general guidelines, and some of them cannot be applied to the Brazilian conditions and rules). These diseases can be differentiated only through laboratory tests and the points below have to be considered during clinical and epidemiological assessment on the premises with suspected animals, pointing out that, in a region where vaccination is not practiced, the clinical picture tends to be much more acute and evident and the attack rate much higher:

- Not always FMD evolves with every classical symptom described; lesions can appear with higher or lower intensity, depending on the virus strain in action, quantity of infective virus and immune status of the animals;
- Cattle are more susceptible to the FMD virus, however, in animals with a certain degree of immunity for FMD, lesions only in their mouths, without generalizing on the feet, or only in one or two feet without oral lesions can occur. An example of this picture was the outbreak in Monte Alegre/PA in 2004, where, at the response to suspected cases, the official veterinary service identified only one cattle with some clinical signs in only one foot. In non-vaccinated herds, the susceptibility does not depend on the cattle age;
- Pigs are more sensitive to the infection and show more severe signs: snout vesicles can be big and full of blood fluid; lesions in their mouth are normally dry with necrotic epithelium; pedal lesions are severe and hooves can be completely detached at the point of the coronary band. The main infection path is digestive, which requires a bigger infectious dosage if compared to cattle. This partially explains the presence of non-infected pigs in holdings with FMD in cattle, as observed in the index outbreak registered in Eldorado-MS in 2005 and during the outbreak in Rio Grande do Sul in 2000;
- In sheep and goats, considering mainly the strains existing in South America, FMD occurs in a more benign way (with mild symptoms) even in non-vaccinated animals. These animals present lesions in their mouth and vesicles in the region of the hoof corona in less quantity, smaller and more difficult to be identified;
- Depending on the strain of FMD virus, not always all susceptible species are affected. In the 2000 and 2001 outbreaks in Rio Grande do Sul, despite the existence of pigs and sheep living together with cattle, only cattle showed clinical signs;
- Swine vesicular disease has a low worldwide incidence, being registered in European and Asian countries, and has never been registered in the Americas. It affects only pigs;
- Vesicular stomatitis is endemic in some regions in Brazil, and the important differential is susceptibility of horses. Therefore, there are cases in which the disease has been identified in cattle and pigs, not being manifested in horses. In cattle, the morbidity rate regarding vesicular stomatitis tends to be higher in adult animals;
- Albeit rare, outbreaks of vesicular stomatitis and FMD can occur simultaneously; thus, even at the concomitant presence of clinical signs in cattle and horses, the possibility of occurrence of FMD (only with laboratory tests) cannot be ruled out;
- Vesicular exanthema has been diagnosed only in the United States and in Iceland. This disease was considered eradicated in 1959 and, since then, other cases have not been registered in any other part of the world.

² Complete viral particle, comprised of DNA or RNA surrounded by protein. It constitutes the last development phase of the virus, that is, the mature infecting particle. Some authors use the term *virion* to identify the viral particle out of the hosting cell.



3.3. *Clinical inspection of animals and epidemiological assessment*

The priority of the veterinarian investigating a suspect case, at the first clinical inspection of the animals, is to confirm or rule out vesicular disease. Irrespective of the susceptible species involved, the verification list must, among other aspects, evaluate the presence of:

- a) high fever of 41°C that decreases after the second day;
- b) Vesicles and full blisters only perceptible during the acute stage of the disease that lasts two days (a vesicle is a small raising of the epidermis containing serum fluid, while a blister is a vesicle bigger than 0.5 cm in diameter, generally made up by the vesicles coalescence);
- c) In dairy herds, strong reduction in milk production before the first clinical signs;
- d) drooling and lameness (pigs particularly show much difficulty in moving);
- e) Bright red secondary erosions, moist and without bleeding, with or without fibrin deposit, in the muzzle, nostrils, mouth, coronary band (corona) of the hoof, interdigital spaces, teats and udder;
- f) sudden death of very young animals caused by hyperacute myocarditis;
- g) Distribution of animals with clinical signs: i) in non-vaccinated species the percentage tends to be high among animals living together in the same pasture, field or barn, which cannot be observed in herds vaccinated several times; ii) in herds with a recent history of vaccination, predominant clinical signs in animals or age groups with low expectation of immune protection; and
- h) Report of the probable beginning of clinical cases with the entrance of susceptible animals in the herd or trucks to load and unload animals. In pig farming, special care must be given to the origin of the feed.

In cases of animals presenting drooling and lameness at the same time, with confirmed or suspect vesicular lesion, measures foreseen for probable cases of vesicular diseases must be taken. In order to maintain the sensitivity of the diagnosis, it is necessary to examine the mouth of each animal limping and the feet of the animals with a lesion in the mouth or in the nostrils.

Confirmation of vesicular disease and the possibility of occurrence of FMD pinpoint another important purpose of the investigation stage: determining the probable onset of the infection. In order to do this, in addition to the information gathered during the interview and history, the detailed description of the secondary lesions that develop after the eruption of the vesicles and the beginning of healing, it is important to estimate the beginning of the first signs and the probable onset of the infection.

Therefore, the definition of “age” of lesions, particularly the oldest ones, is critical to establish the historical evolution of the outbreak, particularly defining the origin of the infection and of the period of higher risk of spreading the viral agent. For this reason, a brochure drafted based on the material published in 1986 by the Ministry of Agriculture, Fisheries and Food of the United Kingdom (MAFF), containing pictures of FMD lesions produced in a study carried out by the Animal Virus Research Institute (AVRI) is part of this document. This brochure also includes pictures of lesions taken in the last few outbreaks of FMD registered in the country. On the OIE website (www.oie.int) there are pictures of lesions of several diseases, including FMD, at link “Animal Diseases Data”/“Technical Disease Cards”.

Once the vesicles are broken, healing speed may be affected by various factors and, the age of the lesion can be approximately estimated. Until the fifth day, it is possible to be exact, with one day error, and after the sixth day, precision decreases as time elapses. Find below some examples to estimate the age of lesions in cattle mouth and in pig feet:

- **unruptured vesicles: up to 2 days**
- **recently ruptured vesicles with pieces of epithelium adhered to the borders of the lesions: 1 to 3 days**
- **vesicles ruptured with epithelium loss and absence of clear borders of fiber tissues: between 3 and 7 days**
- **open lesions with fiber tissue and clear borders: between 7 and 10 days**



3.4. Clinical and epidemiological aspects of other diseases that can be confused with FMD

It should be stressed that the ruling out FMD must be technically well founded and, in case of doubts, expert must continue working considering the possibility of vesicular disease. Remember that, while in the regions without vaccination the clinical picture is more evident, in regions where vaccination is practised, it is not much likely for classic clinical pictures to occur, with easily detectable lesions. In regions where vaccination is practised, the presence of clinical signs is expected in a reduced number of animals with mild lesions, and possibility of being found on the tongue, mouth, interdigital spaces or udder.

In response to suspected cases, the presence of secondary lesions such as erosions, ulcers and scab is the most frequent, although it is not ideal. In this case, the veterinarian must check for some diseases that can confound the diagnosis of vesicular disease: bovine viral diarrhoea/mucosal disease, rinderpest, infectious bovine rhinotracheitis/infectious pustular vulvovaginitis, bluetongue, bovine mammillitis, bovine papular stomatitis, cowpox and contagious ecthyma.

In order to help the veterinarian to clinically distinguish FMD from other classical vesicular diseases, find below some points to be considered, remembering that on the OIE website, in “technical tables” all information on these diseases is available:

- **Bovine viral diarrhoea - BVD:** BVD is caused by a pest virus. This name is due to the fact that the agent was initially identified in cases of bovine gastroenteric diseases. Subsequently, the infection was associated to a large variety of clinical signs, including respiratory, digestive, reproductive, hemorrhagic, cutaneous, and immunosuppression. Although it is associated to different clinical manifestations, most infections of immunocompetent animals by the BVD virus seem to appear without evident clinical symptoms. Due to the epidemiological and clinical-pathological consequences of the infection in pregnant female cattle, the virus is considered important particularly regarding breeding. There must be a suspect of infection of the BVD virus every time there are embryo losses, abortion, fetal malformation, birth of weak animals and prenatal death. In addition, cases of enteric or respiratory disease with hemorrhagic components (melaena, petechiae in mucosa or serosa etc.). Ulcerations in the digestive tract are also signs of infection with the BVD virus. These manifestations can occur isolated, and simultaneous occurrence is a strong indication of the disease. These disorders occur mainly but not exclusively in young animals. In the chronic form, which less common, clinical signs are not specific. Lack of appetite, weight loss and progressive apathy can be observed. Diarrhoea can be continuous or intermittent. Sometimes, there is also persistent nasal and ocular discharge. Areas of alopecia and hyperkeratinization can usually appear on the neck. Chronic erosive lesions can be seen in the oral mucosa and on the skin. Laminitis, interdigital necrosis and hoof deformation can also occur. These animals may survive for many months and they usually die after a progressive debilitation.
- **Mucosal disease:** it is the most severe type of infection of the BVD virus. It occurs in animals that were infected in the uterus and were born with it and remain immunetolerant to the agent. When these animals are overinfected, they develop a severe clinical picture called “mucosal disease” (MD). MD occurs with low morbidity (1 and 2%) and with very high mortality (nearly 100%). It affects cattle between 6 months and 2 years of age and they usually have an acute course. In the acute stage, the disease is characterized by fever (40-41°C), salivation, nasal and ocular discharge, hemorrhagic profuse diarrhoea and ulcers in the nostrils, mouth, eyes, interdigital space with laminitis and coronitis. The ulcerous lesions are found in the whole mucosa of the digestive tract. Laminitis and coronitis can be observed. It is necessary to make the differential diagnosis of the thrombocytopenic form of other hemorrhagic diseases such as acute intoxication with *Pteridium aquilinum*.
- **Rinderpest:** Viral disease of biungulates, caused by a morbillivirus. It can be acute, subacute or chronic with the main lesions characterized by inflammation and ulceration of the whole digestive tract. Occurrence of high fever, anorexia, depression, decreased milk production in dairy cows, ocular discharge, excessive salivation, erosions in the mouth and around it, halitosis, diarrhoea with blood in the stools, severe dehydration, cough and prostration. High percentage of morbidity and death between the third and fifth day after the onset of the clinical manifestations. These are the typical signs observed in infected animals with more virulent variations of the bovine pest agent. Signs must be much milder and some can also be absent with less virulent strains. Different from FMD also for the absence of vesicular and pedal lesions. This disease does not exist in the Americas.
- **Infectious bovine rhinotracheitis/infectious pustular vulvovaginitis - IBR:** It is caused by bovine herpes virus 1 (BHV-1) and affects domestic and wild cattle. After a period of incubation that ranges from 2 to 4 days, the first clinical signs appear such as serous nasal discharge, salivation, fever, lack of appetite and depression. In a few days, the nasal discharge becomes mucopurulent and is followed by conjunctivitis. Abortion and reduction in milk production also occur. The virus can infect the genital tract and cause pustular vulvovaginitis and balanopostitis. Post-mortem

exams reveal rhinitis, laryngitis and tracheitis. Mortality is low. Most infections have a subclinical course from 5 to 10 days. Secondary bacterial infections can cause a more severe respiratory disease. Also observe the absence of vesicular and pedal lesions.

- **Bluetongue:** It is a non-contagious viral infectious disease transmitted by insects of the culicid genus that affects sheep, goats, cattle, deer, antelopes and other artiodactyls. The incidence of the disease is conditioned to ecological factors that control the vector population. In most cases, the disease is not evident, but can cause death (low mortality). The frequency of the infection is higher on cattle than sheep. Therefore, the clinical signs in cattle are rare and very weak when they occur. When visible, clinical signs include fever with inflammation and congestion, oedema on the face, eyelids and ears, haemorrhage and mucosal ulcers. Extended lesions can be found in cheeks and in contact points of the mouth and the molar teeth. The tongue becomes hyperaemic and oedematous, coming out of the mouth, and turning cyanotic in the most severe cases. Hyperaemia can extend to other parts of the body, such as the groins, axillae and perineum. There is also severe muscle degeneration and dermatitis that can be revealed in brittle wool. Sheep can become lame due to hemorrhagic inflammation of the coronary band of the hoof (coronitis) or due to skeleton myopathy. There is a similar disease among wild ruminants caused by the virus of the epizootic hemorrhagic disease that is also an orbivirus but is classified in a serogroup separated from the bluetongue serogroup. When sheep dies due to acute infection, their lungs present hyperaemia and alveolar oedema and the pulmonary tree is full of foam. The thoracic cavity has litres of plasmatic fluid and the pericardial sack shows petechial haemorrhages. Most cases present a characteristic haemorrhage in the basis of the pulmonary artery. In Brazil, the isolation of the virus (serotype 12) was carried out by PANAFTOSA in samples collected by the veterinary service of the state of Paraná during the response to an outbreak in 2001, with 9 cases, and 3 in 2002, with 56 cases, involving sheep and goats. In 1980, the virus (serotype 4) was isolated in the USA from cattle imported from Brazil.
- **Bovine ulcerative mammillitis/bovine herpes mammillitis:** Caused by bovine herpes virus 2 (BHV-2) is clinically characterized by vesicular and ulcerous lesions on the skin of the mammary gland. The disease has a sudden onset, producing oedema and vesicles on the skin of teats and udder. After the vesicles rupture, there is exudate with scabs and ulcerous lesions. These lesions can be localized and discrete, or cover great part of the gland skin. In mild cases, there are discrete nodes or small ulcers, from 0.5 to 2.0 cm in diameter, surrounded by erythema, which is characterized by redness of the skin due to congestion of the capillaries. When it occurs in lactating cows, there can be vesicular and ulcerous lesions in the muzzle, tongue and oral mucosa of the calves. Morbidity can affect 100% of lactating cows, but when the disease is enzootic, it affects only primiparous cows. Typically there are no deaths and the major economical losses result from the reduction in milk production. The diagnosis is made by isolating and identifying the virus in tissue cultures, by electronic microscopy or by the presence of intranuclear corpuscles observed in biopsies of affected tissues. The BHV-2 can also cause a clinical picture of generalized dermatitis, which has not been observed in Brazil.
- **Bovine papular stomatitis:** Viral disease, caused by a parapoxvirus, which can be acute or chronic. In acute cases, the primary hyperaemic outbreaks are spots of 2 to 4 mm in diameter in the muzzle. These lesions develop to dark red and round raised papules. The confluence of various papules turns into a big area with irregular shape. In this stage, the periphery of the lesion becomes red and the centre becomes concave. The lesions are covered by scabs. It is possible to see ulcers on the mucosal surface of the mouth, except the back of the tongue. Some lesions can also be found inside the nostrils. All affected animals recover in 10-14 days. In chronic cases, it is clear the proliferating necrotic stomatitis, profuse salivation, lack of appetite and rising body temperature in addition to necrotic and generalized exudative focal dermatitis covering the whole surface of the body. A clear hyperkeratosis around the mouth, anus and in the ventral face of the tail has also been verified. In this case, mortality is high and occurs between 4 and 6 weeks.
- **Cowpox:** Term used to describe the contagious disease characterized by the appearance of cutaneous lesions in the udder and teats of lactating cows. Three different poxviruses can be involved in its aetiology; cowpox (real pox), vaccinia (virus used in vaccines against pox in humans) and pseudocowpox (pseudopox). The lesions cannot be clinically distinguished. In Brazil, researchers saw this type of sporadic disease from 1950-1970 in the South-eastern states. As of the late 1990s, many outbreaks of a disease similar to cowpox were seen in different regions in the country, such as Vale do Paraíba-SP, in the city of Cantagalo-RJ, and in Mato Grosso do Sul. In the state of Minas Gerais, the first reports of outbreaks date back to 1999 in the "Zona da Mata" region. Some cases have been recently reported in the state of Minas Gerais, Bahia, Rio de Janeiro, São Paulo and Goiás. Information obtained on *SivCont* reveal that 161 outbreaks of the disease were registered from 2005 to 2008, 80 in Minas Gerais, 34 in Mato Grosso, 22 in São Paulo, 10 in Bahia, 6 in Pará, 4 in Tocantins, 3 in Espírito Santo, 1 in Goiás, and 1 in Maranhão. Considering the increase of records of cowpox in the country in the last few years, Appendix 7 has more detail on the disease, drafted with the support of the state veterinary service of Minas Gerais.

- Contagious ecthyma: Also known as contagious pustular dermatitis, infectious labial dermatitis, “crustal” mouth and “chappy lip corner”. Acute infectious disease of viral aetiology that affects sheep and goats, characterized by the formation of vesicles, pustules and crusts mainly on the animals faces. It can also affect humans. The virus that causes the disease belongs to the family *Poxviridae*, Genus *Parapoxvirus*, known as ORF virus. Lambs and kids are usually more susceptible than adult animals. The disease causes weight loss due to oral lesions that makes it difficult for the animal to eat, causing economical loss. Incubation period from 3 to 7 days. At the beginning, an inflammatory reaction in the scarified region can be observed, small vesicles develop and can reach the size of a pea, and present exudation of serum fibrin fluid. In the subsequent 24 or 48 hours, these vesicles are transform into pustules that dry very fast becoming scabs (skin). Lesions are more commonly observed in the lip commissure, but they can also be found on the corners of the lips. In humans, there is usually a single cutaneous lesion, as a chronic cutaneous eruption, circumscripta, very irritating, with tendency to hyperplasia.

Information on the characteristics of the lesions and susceptibility of domestic species and human beings in vesicular diseases is summarized on Table 1. In addition, Table 2 summarizes the main characteristics of FMD and vesicular stomatitis, as well as the most common diseases diagnosed in the surveillance system for vesicular diseases in Brazil.

In addition to the infectious diseases that can be confused, other common cases of ruling out suspects of vesicular disease involve intoxication and physical or chemical traumas. In case of intoxication, the substances that cause photosensitivity, caustic or abrasive chemicals, and fungi *Phytomyces Chartarum* and of the *Clavaria* genus must be highlighted.

The *Phytomyces Chartarum* fungus cause facial eczema and affects cattle and rarely sheep, characterized by a clinical picture of photosensitivity.

Fungi of the *Clavaria* genus are related to eucalyptus crops, hot seasons with high humidity. It is more common in the Southern Region of the country and the clinical picture observed in cattle and sheep is called BOCOPA (short for Spanish *Boca-Cola-Pata*) in Uruguay. Infected cattle can neither drink nor eat and swallow food, presenting intense salivation and, at mouth inspection, particularly the tongue, total detachment of the necrotic epithelium. Congestion of conjunctiva can be observed and, in sheep, opacity of the cornea may occur, causing blindness, in addition to lameness and fall of the animals that cannot stand up. In cattle, slackening of the horns that are loose very easily is observed. Shedding of tail hair is constant. Among laniferous animals, it is possible to observe shedding of wool after a few days. Depending on the quantity of fungus eaten, the animals may die after a few days.

With regard to traumatism, different elements can cause salivation and lameness, particularly lesions caused by dry and hard pasture, recently cut pasture and crops (tuft of grass and second cutting) and by soils with predominance of gravels. In dairy cattle farms, pedal lesions are common, and the following may be observed: interdigital dermatitis; heel horn erosion; heel erosion; verrucose dermatitis; vegetative interdigital pododermatitis; digital dermatitis; interdigital phlegmon; diffuse aseptic pododermatitis; pododermatitis circumscripta; necrotic pododermatitis; claw fissure; white line disease; fracture of phalanges; sole and heel abscesses; sole ulcer; toe ulcer; increased arthrosis; dislocations; and sole haemorrhage. Foot rot is another common lesion in sheep farming.

Table 01. Susceptibility and characteristics of lesions for vesicular and confoundable diseases

Disease	Species						Main characteristics of the lesion
	Cattle	Pigs	Equine	Sheep	Goats	Human	
Foot-and-mouth disease	+++	+++		++	+		Vesicle
Vesicular stomatitis	+++	+++	+++	++	?	+	Vesicle
Swine vesicular disease		+++	?				Vesicle
Vesicular exanthema		+++				?	Vesicle
Bovine viral diarrhoea	+			?			Ulcer
Malignant catarrhal fever	+			?	?		Papula or erosion
Infectious bovine rhinotracheitis	+						Ulcer
Bovine papular stomatitis	+					+	Papula
Contagious ecthyma	+			+++	+++	++	Crust and pustule
Bluetongue	++			+++	+		Papula
Herpes Mammillitis	+	+		+			Vesicle Ulcer
Rinderpest	+	(+)			+		Necrosis ulcer vesicle
Cowpox	+++					+++	Ulcers and Crusts

Source: Adapted from the Manual of Procedures for the Response to Foot and Mouth Disease and other Vesicular Diseases, PANAFTOSA

Symbol “+” indicates the susceptibility of the species, the more symbols there are, the greater is the susceptibility. Symbol “?” indicates uncertainty as to the species’ susceptibility.

Table 02. Characteristics of FMD, vesicular stomatitis and main diseases involved in the veterinary surveillance system for vesicular diseases in the country.

Characteristics	FMD	Vesicular stomatitis	Bluetongue	IBR	BVD
Morbidity	High (60 to 100%)	Low to medium (5-10%); in dairy cattle herds up to 85%	Medium to high – depending on the presence of vectors (50-75%)	8% (Milk) 20 to 100% (Fattening)	BVD – Low to Medium Mucosal disease (5-10%)
Mortality	Low (it can be high in young animals)	Zero or low	20 to 50%	0 to 3% (Milk) 1 to 10% (Fattening)	BVD – low to medium (sporadic; 0-20%) Mucosal disease (90-100%)
Transmission	Contact, aerosol, meat products. Uncertainty about the carriers’ role. Windborne only under special conditions (temperature, humidity, pressure, wind)	Uncertainty about the role of contacts, carriers and vectors. Milking machines	Vector (<i>Culicoides</i> spp.) Uncertainty about the role of cattle as carriers.	Carriers Contact/Aerosol Coitus/Semen	Contact Persistently infected cattle Important vertical transmission
Notes	Persistence in cattle. Virus in faeces, urine, milk, oesophageal-pharyngeal fluid, breath and lesions. Considered the most contagious disease in veterinary medicine.	Calves are more resistant than adults. New Jersey serotype is more virulent than Indiana. Zoonosis. Natural immunity < 6 months. The virus does not survive more than 1 or 2 weeks in the environment. Fibrous Food exacerbates the infection/ transmission. Wild fauna?	Carrier cattle (?), Reservoir (?) Vector seasons. Quite resistant to the environment. Differences in the susceptibility according to breed and age (lambs are more resistant)	Persistent infections – reactivation (with stress?) Wild animals can have an important role in Africa. Vaccination provides protection – 9 months Protection by colostrum ranges from 1 to 6 months	Isolation of virus in faeces, urine, saliva, semen, milk. Congenital infection is important in persistent BVD
Necropsy	Not carried out. Evidence of erosion in the rumen – white signs/streaks in the myocardium (tiger heart)	Not carried out.	(Not evident infection of a traumatic pathology) Abrasion in the mouth/ nasal/ palatal mucosa (mainly petechiae and ecchymosis) – in pleura and serosa. Also in the mucosa of the digestive tract, heart, bleeding enteritis, facial oedema, aspiration pneumonia	Airways: Erosion of nasal cavity, palate, trachea, bronchi Digestive Tract: Similar to Rinderpest – thick exudate that covers the ruminal mucosa – grey. Focal areas of necrosis. IPV/BP – Blanched pustulas in the genital mucosa (pustular vulvovaginitis and balanopostite)	Stomatitis/Catarrhal Pharyngitis – with necrosis spots Dehydration Bleeding Enteritis, lymph gland oedema. Erosion of ulceration in the digestive tract. Mucosa – hyperaemic to hemorrhagic w/oedema. Diphtheric membranes. Lysis of the lymph nodes. Hyperplasia of the cerebellum.

Source: Adapted from the Manual of Procedures for the Response to Foot and Mouth Disease and other Vesicular Diseases, PANAFTOSA

Obs.: words or sentences followed by the interrogation mark indicate uncertainty on the subject.

3.5. *The importance of rapid notification, rapid response and information records*

One of the main indicators of efficiency of the veterinary surveillance system is the number of responses to suspected vesicular disease carried out by the official veterinary service. The lack of response can mean both a real lack of occurrence of the clinical signs compatible with vesicular diseases and a lack of motivation or preparedness of the local community for communication of notifications, affecting the quality and the credibility of the surveillance.

The notification to the veterinary service may be effected by the owners or people responsible for the animals, other representatives of the community (special attention to private veterinarians) or as a result of the surveillance carried out by the official veterinary service. Therefore, in summary, the types of notification are grouped as **surveillance**, when the communication starts from the representative of the official veterinary services, **owners**, when it starts from the owners or people responsible for the animals, and **third parties**, when it starts from any other representative of the community who is neither the owner nor responsible for animals. The last two express the participation of the community in the process and are part of the passive surveillance system. The larger the number of notifications by the owners, the better the surveillance system (high sensitivity), because it proves the close relationship between the community and the official veterinary services by the effective participation and commitment of all. This indicator is therefore directly related to the activities of sanitary education.

People in charge of the local veterinary units and their team must promote educational activities, such as meetings and seminars, in order to motivate farmers. Sanitary education activities must also be indirectly provided to farmers, particularly rural schools, with the participation of the local teachers and their students. The purpose of these activities is to inform the community on the main procedures to prevent the introduction of the disease and the clinical signs indicating its occurrence (*drooling and lameness*) stressing the importance of rapid notification. Also, in order to do a timely notification, the community must trust the official veterinary service specially regarding the guarantees of possible compensation for animals and products that would be destroyed. Special importance must be given to the role of the private veterinarians and agricultural technicians. These experts must be included in primary prevention works (including educational activities) and, depending on the dimension of the sanitary action and on available human resources in the official veterinary service, they must be hired or temporarily called to help in disease emergencies, after being trained.

The other way for a notification to be made to the official veterinary service is **surveillance**, which is the result of investigations made by the official service within the active surveillance system. Disease identification by active surveillance represents, on one hand, the good investigative capacity by the official veterinary service and, on the other hand, the low participation of the community. The most common is that, after confirmation of the disease through a notification made by the community, other cases are identified through the investigation carried out by the official veterinary service (this is observed in the outbreaks in Rio Grande do Sul, 2000 and Mato Grosso do Sul, 2005). Other examples of notification are the communications of employees of a certain local veterinary unit that have been notified of suspected disease in a holding located in a city under the coordination of another veterinary unit, or information gathered from retailers of veterinary products particularly regarding sales of large quantities and to a certain producer, of products used for disinfection and disinfectant foot bath.

Another critical element for success of the emergency actions is recording the time elapsed from the onset of the occurrence to its notification, between notifications, and the first actions of the official veterinary service. Time is the main “enemy” to be overcome by the official veterinary service, especially in regions where vaccination is not practised. Operations during a veterinary emergency aim to quickly control and eliminate possible sources of infection, considering that, the shorter the intervention time, the lower will be the losses from the disease spreading. The time frame to evaluate the effectiveness of the animal health system described above is the following: i) *action time* → the time interval between the probable onset of the disease and notification; and ii) *response time* → time interval between the notification and the visit by the official veterinary service. The *action time* evaluates the degree of participation, knowledge and commitment of the community, while the *response time* evaluates the capacity and preparedness of the official veterinary service.

According to the standards, owners, veterinarians and other community representatives have up to 24 hours to notify any suspected case of vesicular disease to the official veterinary service and, the veterinary service has 12 hours to respond (art. 4, Normative Instruction nº 44, of October 2nd, 2007).

When a diagnosis of FMD or any exotic disease is confirmed in the country, DSA/SDA/MAPA has up to 24 hours to communicate the occurrence to the OIE, including also the communication to PANAFTOSA, neighbouring countries, economic blocks and other commercial partners of Brazil.

But, in order to have an appropriate evaluation of the surveillance system, the existence of suspected cases and the rapid response to them are not enough. The entire response must be correctly recorded by the official veterinary service.

Each and every action executed by the official veterinary service has to be recorded and proven through specific documentation (that is, it can be checked during an audit), otherwise the activities cannot be considered. On the other hand, only through the record of the action, will it be possible to quantify the indicators described and to identify the efficiency of the animal health system.

Professionals in local veterinary units must be prepared for and aware of the need to correctly record every activity carried out, including false accusation, traumas, poisoning, foreign bodies, and other cases where suspected vesicular diseases were ruled out.

For correct registration of the information concerning response to suspected cases of vesicular diseases, the following means must be used:

- **Log of sanitary events:** In every local veterinary unit, there must be, under the responsibility of the official veterinarian, a logbook for registration of every response to the notification of suspected vesicular disease (this book can be replaced IT systems). The minimum information for registration is detailed on item 3.6 of this document.
- **FORM-IN (Disease Investigation Form – Initial) and FORM-COM (Disease Investigation Form – Supplementary):** FORM-INS are forms used by the official veterinary service that must be filled in even in cases of suspected cases ruled out right after the first visit. The official service veterinarians must be prepared to correctly fill in each field in the forms in accordance with the specific guidelines of the instructions made available by DSA/SDA/MAPA.

In the local veterinary units, the forms must be organized in a specific file. The organization must be done by response given, considering the chronology of the facts. The veterinarian in charge of filling in the forms (FORM-IN and FORM-COM) must forward them immediately to the central unit of the state veterinary service when a suspected likely case of vesicular disease is confirmed or together with the weekly information when the suspected case is ruled out. Remember that when there are samples collected for shipping to the laboratory, the samples must be accompanied by the respective FORM-IN or FORM-COM (this last one in case of supplementary visits). The central unit of the state veterinary service must immediately send copies of the forms to the corresponding Federal Superintendency of Agriculture (SFA) and to DSA/SDA/MAPA.

- **SivCont (Continental Epidemiological Surveillance System):** computer web-based system developed by PANAFTOSA, in response to the request of South American countries, and adopted by Brazil as of the second half of 2004. All information recorded in the previous media must be used to feed *SivCont* which, in addition to providing more transparency to the national system of veterinary surveillance, enables the correct filing and fast compilation of data. In Brazil, DSA/SDA/MAPA, with the support of PANAFTOSA, provides training to the state veterinary services and produces and provides specific manuals. The central unit of the state veterinary service, based on the forms sent by the local veterinary units, is responsible for immediately entering and updating the data on *SivCont*. SFA follows-up on the events entered in the system.
- The registration of other actions regarding the surveillance system, such as visits to holdings, must be made according to the guidelines of the *Manual for Surveillance of Vesicular Diseases – CFA/CGCD/DSA*.
- We recommend that the state veterinary services provide the local community with a free system to notify suspected animal diseases, such as hot lines and e-mail. We also recommend that the veterinary services provide the local community with brochures and guidelines on the disease, including the typical signs and procedures to notify suspect cases. (making them available to the farmers in case of ruled out suspect responses).

3.6. Step by step in the response to and investigation of notifications of suspected vesicular disease

Find below the procedures to be adopted by the official service veterinarians in case of notification of suspected vesicular disease. We have to highlight that, in addition to the importance of the response time, another critical action is the correct and thorough registration of the activities carried out, as specified in the previous item.

After receiving a notification, the veterinarian must:

1st Immediately record the notification of the suspect in the logbook of sanitary events

At first, the records must include at least the following:

- Date and time of the notification;
- Type of notifier (owner, third party or surveillance);
- Name of the notifier (when relevant, include the nickname) and contact telephone;
- Identification of where animals with suspected vesicular disease are located;
- Identification of the employee that received the notification;
- Short description of the notification (include the species involved);
- Other remarks considered to be relevant; and
- Date and time of exit for response.

If the notifier does not want to be identified, total assurance of confidentiality must be given. In this case, item “identification of the notifier” must be filled in as “anonymous”.

When the notification is made by telephone, we recommend as a precaution that the telephone number is written down in order to call back to confirm. However, if the person does not want to be identified, confidentiality must be guaranteed.

If the person that notified the suspect was in contact with the animals, he/she must be very well informed on the necessary biosecurity procedures to prevent the spread of the possible infectious agent, particularly with regard to not moving sick animals, and their direct contacts, and not to enter any holding with susceptible animals until the result is given by the official veterinary service.

If the professional of the state veterinary service is out of his office when notification is received, it is important that, before going to the farm, he/she goes to the office to take the measures and recommendations set forth in this manual. If the veterinarian is near the premises where the suspected cases of vesicular disease are, he/she must make a **preliminary assessment**, provided that biosecurity measures are ensured and the activities are recorded.

2nd Initial Gathering of Information

At first, data available on the registration system or in the farm records at the state veterinary service must be evaluated such as the existing herd; quantity of animal movement (especially entrance and exit in the last 30 days); date of the last vaccination; geographic location and roadways. Also identify the neighbouring holdings and the related holdings (that had some relationship – entrance/exit – with the holding with animals under investigation in the last 30 days). Also gather information on other holdings belonging to the same owner. If the holding is not registered in the local veterinary unit (take the opportunity to include it in the records), it is necessary to find its location and identify the neighbouring holdings. This first information gathering must be made rapidly and objectively so as not to affect the response time. The priority is to actually visit the holding. Depending on the result of the response, new data must be obtained in order to carry out supplementary analysis.

Before leaving to visit the holding for the first time, the veterinarian in charge of the local veterinary unit must inform the team to wait for him/her to come back and for the result of the notification before issuing GTAs with origin in the suspect holding and in the holdings related to it, for both geographic proximity or animal movement.

3rd Travel to respond to a notification

In parallel to the initial information gathering, the vehicle to drive to the farm and the response kit for suspect vesicular diseases must be prepared. The team and the superior must be immediately informed about the time and reason for leaving: response to a notification of suspected vesicular disease.

Response must be preferably immediate or occur within 12 hours. In case of notification by third parties or by surveillance, try to identify and contact the owner or person responsible for animals in advance to agree on the best and quickest way to carry out the clinical inspection of the suspected animals. If the notification was made toward the end of the day, and depending on the distance and on the conditions of the highways and lighting at the holding, we recommend that the visit is made in the first hours of the following morning. If the veterinarian in charge of the local veterinary unit is not present at the moment of notification, the employee that receives it must contact the central unit or the regional unit (if there is one) to evaluate and decide on the visit being made by another official veterinarian. In case of resistance by the owner or the person responsible for the animals, the notification must be assisted by the police, but all measures must be taken before taking this action. The professionals of the official veterinary service must bear the official identification card or another professional identification document and a copy of the legislation that gives them competence to take the necessary measures regarding animal health particularly entering the holding or other premises to examine the animals suspect of being sick and blocking the premises if the risk of the infectious disease being present and spreading is confirmed.

The mandatory document to record the first activities is the FORM-IN. Filling it in correctly is extremely important to the epidemiological assessment and to keep the information system updated on the response to the disease events. **We recommend** that the professional brings to the holding only the copies of the FORM-IN to take notes, filling in the final version after going back to the office. It is important that the official service veterinarians understand and know how to **appropriately** fill in each field in the form. And for that matter, the instructions to fill in the forms published by DSA/SDA/MAPA must be read **in advance**.

Response and investigation of the suspected case must be made as quickly as possible. Therefore, the Professional must go directly to the holding with the suspect cases, and not stop in other holdings on the way. If the notification was made on holidays or weekends, the person in charge of the response must have complete autonomy to use the vehicles and all the infrastructure of the institution required to carry out the work.

4th Actions on the holding

Once arrived on the holding with suspect animals, the professional must take every care with the biosecurity and must be dedicated to the investigation works, interview and clinical inspection of the animals. Some points to be considered:

- in the case of small holdings, the vehicle used should remain at the entrance. If the holding is too big, it is necessary to go directly to the farmhouse to carry out an initial interview with the people responsible for animals and to define the best way to carry out the clinical inspection. In bigger holdings, drawing a simple map, indicating the location of the pens or pastures and the distribution of the susceptible animals, is very helpful for epidemiological assessment;
- when entering on the holding, the professionals must wear appropriate overalls and boots;
- go, with all necessary material, directly to the batch of animals with suspected FMD and inspect them, if possible in the same place where they are. If necessary and if the risks for spread of the disease are reduced, the animals may be relocated on the holding to a place where clinical examination can be done. The inspection must start with the suspect batches, given that in this stage of investigation, the most important thing is to confirm or rule out the suspected infectious vesicular disease;
- In the batch under investigation, inspect as many animals as possible. For transmissible diseases such as FMD, the order of inspection of the animals that live together is not epidemiologically important, it does not matter whether one starts with the healthy animals or with the animals with clinical signs. However, considering the need to rapidly evaluate the suspected case presented and, especially in situations in which clinical inspection of the animals is complex, we recommend that the inspection starts with the animals showing clinical signs in order to collect material. It is important to examine as many animals as possible (do not forget to use gloves), both with clinical signs and appearing to be healthy, to evaluate the spreading of the disease, the age of lesions and to establish the probable onset of the outbreak based on the interviews.

The veterinarian in charge of the response must keep in mind that, depending on the clinical and epidemiological picture found, other visits may be necessary for supplementary inspections in the herd. The first visit has the priority of ruling out or confirming the suspected case and, when necessary, sampling for shipping to the laboratories of MAPA. In Appendix 8 there is a basic guide to be consulted, for examination of animals with suspected vesicular diseases;

- In addition to clinical inspection, an epidemiological assessment must also be carried out considering the indicators of animal demography (age, sex, density, type of production etc.), expectation of immunity of the existing animals, recent entrance of animals in the lot, changes in animal handling, simultaneous occurrence in different species, pasture and soil quality (if there are tree stumps or stones, etc).

The clinical investigations carried out at first (still on the holding) are useful to underpin the analysis of the health status of the animals, guiding the veterinarian to establish a final or temporary diagnosis that will result in: ruling out/confirming the suspect of vesicular disease.

a) Ruling out vesicular disease

Ruling out on the holding must occur in the following cases:

- Non occurrence of any disease, such as in case of false accusation;
- Occurrence of a non-infectious disease (poisoning, foreign bodies, traumas); or
- Occurrence of another infectious disease presenting a clinical and epidemiological picture incompatible with vesicular disease.

The veterinarian must analyze all the information that founded the diagnosis, recording it on the FORM-IN.

Where vesicular disease is ruled out, when other transmissible infectious diseases are suspected, sampling for laboratory confirmation of the diagnosis must be done, preferably of material for isolation and identification of the etiological agent (fragments of organs, lesions and crusts, lesion swabs, etc.). This includes cowpox, IBR and BVD in the clinical stages where the signs are separate from the typical vesicular lesions. In suspected cowpox, always remember to wear gloves to collect samples. The samples must be sent to the laboratory, with the corresponding FORM-IN (the FORM-IN must be filled with the presumptive diagnosis such as: **cowpox, contagious ecthyma etc** – do not use the term “vesicular disease” in these cases). Sampled animals must wear permanent identification. The laboratory must be part of the LANAGRO network (if it does the tests involved) or among others with the capacity for desired diagnosis. The FMD Coordination is the sector of DSA/SDA/MAPA responsible for maintaining updated the list of laboratories for diagnosis of vesicular and other diseases that can be mistaken for it.

Still with regard to other suspected transmissible non-vesicular diseases, once leaving the holding biosecurity procedure must be adopted; direct return to the local veterinary unit is recommended.

If no other infectious diseases are suspected, the investigation must be concluded in the FORM-IN, recording on it the final diagnosis issued and the information that underpins it. In these cases, it is not necessary to fill the conclusion FORM-COM.

In all cases, take advantage of the visit to update the existing records information and the herd information. If the holding is not registered in the state veterinary service records, take the information necessary to include it in the database and give to the owner or responsible for animals the guidelines and information on the legal and sanitary aspects involved.

b) Confirmation of a suspected probable case of vesicular disease

In case vesicular disease is confirmed, the veterinarian must give special attention to the following activities: *sampling of material for diagnosis, information gathering and biosecurity*. Some procedures and information on each activity to be considered at the place where the probable cases of vesicular diseases occurred will be highlighted. We must highlight that, between the confirmation of the suspected case and the result of the laboratory tests, there is a very important time interval when the possibility of there being FMD must be considered. Depending on the quality of the sampled material, after arriving in the laboratory, the initial result can be released in less than 24 hours.

Sampling of specimen for diagnosis:

The impossibility of a clinical differential diagnosis of vesicular diseases, associated to the frequent lack of epidemiological information at the beginning of the investigation, needs the laboratory support for confirmation of the diagnosis. For a correct sampling, the following must be highlighted:

- every animal submitted to sampling must present permanent or long-duration individual identification that must be used in the identification of the flasks with samples;
- the chosen material is made up of fragments of vesicular epithelium, including the borders of the lesions. If the vesicles are entire (not broken), in addition to the epithelium, there must be also a



Vesicular fluid that can be sent to the laboratory inside the same syringe used for sampling, being duly sealed, identified and kept refrigerated;

- the material collected from the oral and nasal regions is more appropriate when not dirty. The feet and udder, before sampling, must be washed with clean water to remove dirt (do not use any kind of soap or antiseptics). Put the collected material inside separate flasks, containing the *Vallée Medium*, for each animal involved. The *Vallée Medium* contains a buffer solution of phosphate glycerin, the main function of which is to maintain the pH stability, that must range from 7.4 to 7.8. This preservative must be maintained between 4 and 6°C (formula available on Appendix 10). It must have a pH indicator or be tested before its use.
- keep the material collected from the oral and nasal regions in separate flasks from the material collected from the feet and udders. Collect at least one gram of material (the quantity sufficient to complete an imaginary square of 1 to 2 cm²). If necessary, in order to complete the minimum quantity of samples, it is possible to put in a single flask, fragments of epithelium of the oral and nasal region, with fragments of the feet and udder, but never mix the material of different animals in the same flask. Add the preserving fluid in a quantity sufficient to cover the whole material. The flasks must be duly sealed, identified (same as the animal identification) and kept refrigerated or, better, frozen (-20°C). After being sealed, the external part of the flasks must be cleaned and disinfected before being placed in the isothermal box (the use of small sprayers or manual squirters, with disinfecting solution, facilitates this operation);
- evaluate the animals in different stages of the diseases, trying to establish the age of the lesions, as described in item 3.1. This is an important point, in which the veterinarian must evaluate the quantity of animals for inspection. In case he finds new cases, that can be easily sampled the professional must increase the number of inspected animals (but without affecting the response time) with the purpose of detecting the oldest lesions to support the definition of the probable onset of the disease. However, if he finds only old lesions that are difficult to sample, the professional must inspect as many animals as possible, with the purpose of finding more recent lesions, with more possibility of virus isolation;
- the owner or person responsible for animals must be told not to treat the animals in order not to affect further collection of samples, if necessary;
- undesired situations, but that can be found on the holding, are of animals treated with a certain type of Antiseptics or of animals with old lesions, without much possibility of epithelium sampling. Both situations represent a deficiency of the surveillance system (the veterinary service arrived “late”) and have to be corrected. In these situations, it is necessary to insist in the collection of the epithelium of an oesophageal-pharyngeal fluid (OPF), using an appropriate cup, in accordance with the PROBANG technique. These situations must be registered on the FORM-IN. Procedures for collection of OPF are described in Appendix 9, underlying that the veterinarians need the specific training for realization of this activity.

The OPF collection needs specific training and the animals must have fasted for at least 12 hours. It must be necessary the return to the holding on the following day to make the collection. The collected fluid must be stored in the same quantity of the Earle Medium (formula available in Appendix 10) and frozen as rapidly as possible. This medium contains antibiotics, fungicides, and different enriching products to preserve the collected cells. A sample of OPF can be considered negative when the cytopathic effect is verified after at least 3 passages in tissue culture and when negative on the complement fixation test. Virus isolation from the OPF presents low sensitivity. In case of negative samples, it is recommended to take one or two more samples, with an interval of at least 15 days, as an attempt to obtain a more consistent diagnosis. As it can be seen, the collection of oesophageal-pharyngeal fluid (OPF) in suspects of FMD is not an ideal technique, therefore it should be used only as a last resource.

Vallée and *Earle Medium* used to preserve epithelium samples and oesophageal-pharyngeal fluid (OPF), respectively, have different compositions (Appendix 10). In addition to preserving, these media have the purpose of preparing the samples for various procedures which they must be submitted to in the laboratory. Thus, the use of these media must respect the specific aims for which they have been prepared, that is, it is not recommended to replace one for the other.

In these situations, where the lesions are old or were treated, another possibility is to collect blood serum. The OIE, through the guidelines for surveillance of FMD, in the item referred to serologic surveillance, describes that a positive reaction to the test of detecting antibodies against the FMD virus, must have four causes: Natural infection; vaccination; presence of maternal antibodies; and cross

reactions (heterophyllous). The sensitivity and specificity characteristics of the laboratory test used, as well as the positive predictive value and the negative predictive value of the results found must also be considered³. With regard to maternal antibodies, the OIE also points out that, in cattle, these antibodies are usually found only at six months of age, but that in some individuals, they can be detected for longer. In Brazil, professionals in the vesicular disease diagnosis have frequently registered the detection of maternal antibodies in cattle older than six months.

In regions where vaccination is not practised, the identification of antibodies against the FMD virus is an easier to be analyzed, but it always has to be linked to the clinical and epidemiological picture found. As presented in item 3.1 of this document, in an animal without vaccination history, the production of humoral antibodies begins after the 5th day of infection, reaching detection levels after the 14th day with the available laboratory tests. In this moment, the healing process is in progress and the possibility of virus isolation is small or nonexistent. Specifically in herds without vaccination history, considering the transmission characteristics of the viral agent and depending on the production system and on the animal concentration levels involved, a high incidence rate (attack rate) is expected. Considering the region involved, it is also expected to detect other infected herds, and the speed and coverage of the spreading of the disease depends on the characteristics of the predominant production system, concentration of holdings with susceptible animals, in addition to intrinsic factors of the viral agent. Therefore, despite the identification of antibodies against the FMD virus, in animals with clinical signs of vesicular diseases, representing a conclusive finding in non-vaccinated herds in areas where there is an implemented surveillance system is more probable than the confirmation of the case by virus isolation and identification. In any case, in this initial phase of the investigation, the serum sampling must be limited to the animals with clinical signs, also because these samples can also be used in the support to the differential diagnosis, as detailed below.

Where vaccination is not practised, the use of laboratory tests to detect antibodies against the FMD virus has a limited value when analyzed individually. Possible situations involve the identification of non-vaccinated herds or of group of non-vaccinated animals in a herd with a history of vaccination in the region under investigation. In these cases, the incidence rate tends to be high among the animals with low immunity. The veterinarian in charge of investigating the suspect of vesicular disease must pay special attention to the history of vaccination against FMD, try to cross-reference the information and attempt to find material evidence. As it was mentioned in the previous case, in this stage of investigation, blood serum sampling must be limited to the animals with clinical signs. With regard to the animals with a vaccination history, the purpose of these samples must only be to support the differential diagnosis. In some cases, after the analysis of the age composition of the herd, it could be necessary to go back to the holding to take more samples, defined as a static basis, to evaluate the possibility of circulation of the FMD virus.

With regard to the epithelium samples and oesophageal-pharyngeal fluid (OPF), Figure 4 brings a graph of the diagnostic laboratory, drafted based on LANAGRO/MAPA data and on PANAFTOSA Training Manual n° 15. It is highlighted that, when the quantity of samples is appropriate, the result can be obtained in less than 24 hours. On the contrary, the laboratory tries to “improve” or “enrich” the sample sent through the passages in culture medium of inoculation in mice, which could last 10 days.

³ Sensitivity: the likelihood of an animal being classified as positively infected in a test.
Specificity: the probability of an uninfected animal being classified as non-infected in a test.
Negative predictive value: proportion of negative animals which are not infected.
Positive predictive value: proportion of animals positively infected.

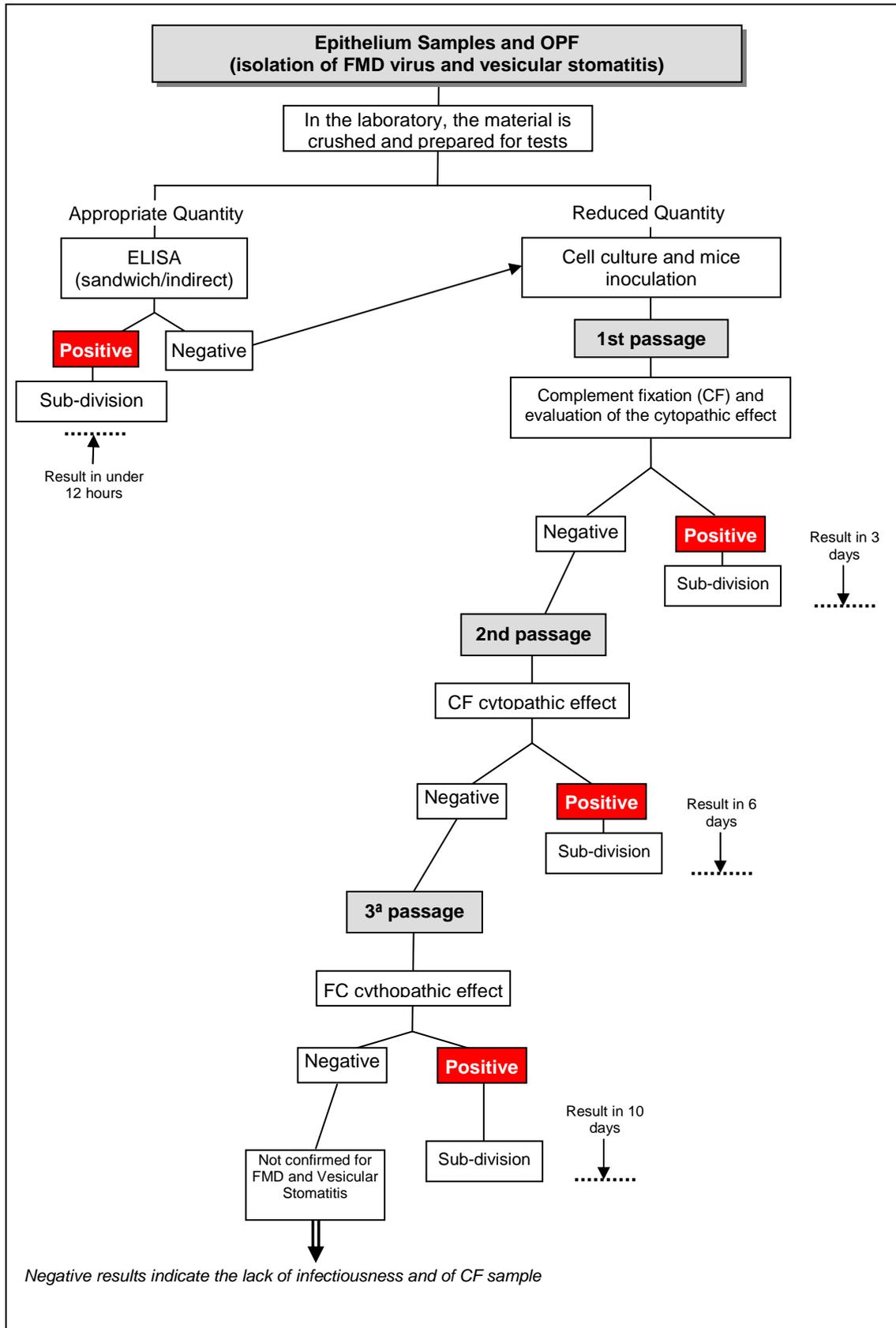


Figure 4. Simplified flow of the epithelium samples in the laboratory for diagnosis of FMD and vesicular stomatitis

Taking into consideration the differential diagnosis, it is important to include the material below in the collection, in accordance with the PANAF-TOSA *Manual "Procedimiento para coleta y remisión de muestras para el diagnóstico de enfermedades vesiculares y su diagnóstico diferencial"*. It must be highlighted that the tests for differential diagnosis must be made after the negative result for FMD and vesicular stomatitis. Depending on the quality and quantity of samples collected during the first inspection, it must be necessary to go back to the farm to collect new samples.

- Secretion swabs and lesions: For isolation of the IBR and BVD virus, the material must contain epithelial cells. In order to do this, we recommend to harshly scrub the ocular or nasal mucosa and put it immediately in an appropriate tube containing EAGLE MEM medium with 10% bovine fetal serum and the double amount of antibiotics (composition in Appendix 10). If BVD is suspected, the ideal is to take serous secretion samples and not mucopurulent. If IBR is suspected, oral, anal, vaginal or preputial swabs must be made.
- Total blood with EDTA or Heparin (1 mg/ml): quantity of 3 to 5 ml per sample, that cannot be frozen. It has the purpose of supporting the diagnosis of BVD and Bluetongue.
- Blood serum: in the initial stage of the investigation, the samples of blood serum must be limited to animals with lesions. If necessary, blood serum sampling can be taken from other animals on the holding, which will depend on a subsequent epidemiological assessment to determine the species, age groups and quantity of samples involved. Taking blood serum samples, after 20-30 days to evaluate antibody titers might be necessary. The material to be sent to the laboratory must not be haemolysed blood serum (at least 2 ml of serum/animal). In order to obtain 2 ml of serum, collect at least 10 ml of total blood.
- Organs: depending on the disease, the following samples can be shipped frozen:
 - IBR: Fragments of mucous membranes of the respiratory tract, tonsil, lung and lymph nodes. In case of abortion, take samples of placental cotyledons and of liver, lung, spleen and foetus kidneys. In case of detection of nervous signs, send the cephalorachidian fluid.
 - BVD: samples of small intestine, Peyer's patches (agglomeration of lymph nodes located mainly in the ileum mucosa), oesophagus, lung, adrenal, mesenteric lymph nodes and fetal tissues.
 - Bluetongue: Spleen, liver, bone marrow, heart blood, lymph nodes and ulcerated mouth epithelium.

Information gathering (epidemiological investigation):

After the verification of the possibility of vesicular disease, information gathering must be deepened through an interview with the owner or with the people responsible for the animals. The questions must help determine the probable date of the onset of the disease event, the possible origin and to evaluate the risk degree of its spreading. In order to do this, remember that, the incubation period of FMD is maximum 14 days, but 2-7 days is more common.

The veterinarian in charge of the response must meet the people directly involved, interview them and provide them with all instructions and recommendations. Check if all information to fill in the FORM-IN is available, and, if necessary, finish it at the office. Special attention must be given in filling in the field for the probable onset of the disease, comparing the results of the interview and the evaluation of the age of the lesions examined during the visit.

Verify the entrance of susceptible animals and vehicles, particularly trucks that carry animals or animal products, at least in the last 30 days compared to the onset of the first cases of the diseases. Verify also the existence of a possible relationship of the owner or person responsible for animal handling with other herds located in Brazil or other countries.

Check the recent presence of professionals such as veterinarians, agronomists, agricultural technicians, inseminators, vaccinators, animals dealers, among others that had contact with susceptible animals.

Biosecurity activities:

As already mentioned, the period between the confirmation of the clinical suspect and waiting for laboratory result is very important, and the possibility of it being FMD must be considered.

In areas where vaccination is not practised, the disease can spread very fast among susceptible animals. In most cases, transmission occurs after direct contact between infected and susceptible animals; a large quantity of virus is found in all secretions and aerosols 1 to 3 days before and 7 to 14 days after the appearance of lesions (in vaccinated animals, elimination of the viral agent before the appearance of lesions has not been found). Less frequently, the virus is mechanically spread among infected and susceptible animals through animal products, instruments (fomites), vehicles and people.

Biosecurity measures are a set of activities used to prevent or minimize the risks of the disease spreading, and several procedures must be followed on the holding where the suspect was confirmed:

- After taking the samples, gather all disposable material, put them in plastic bags and clean and disinfect the remaining material, that must also be appropriately stored;
- Go to the place where the interview will be made with the owner or the person directly responsible for handling the animals; change clothing, put away overalls and boots in appropriate plastic bags;
- Draft the Notice of Blocking and clearly and objectively communicate the guidelines and care to be taken to prevent the spread or aggravation of the sanitary problem. The local veterinary units must have Notice of Blocking and Notice of Blocking Lifting forms for prompt use. The Notice of Blocking must contain the reason for its use, its legal basis, a field for the signature of the owner or the person responsible for the herd and the main prohibitions established (templates available in Appendices 11 and 12);
- Among the main guidelines and prohibitions that must be followed, considering the size of the holding and predominant livestock production system, the following items can be highlighted:
 - Prohibit the exit of animals and products with risk of spreading FMD from the holding. Non-susceptible animals should be included considering the risk of mechanical spreading of the disease;
 - Products that are not directly linked to the risk of spreading the disease can spread it mechanically, and all measures to disinfect vehicles and packaging of these materials must be taken;
 - Stop works with tractors and machinery that could increase the chances of mechanical spreading of the disease;
 - Leave the lot in cases of disease under the responsibility of only a small group of workers that should not have access and contact with other susceptible animals on the holding;
 - Instruct everyone not to visit other holdings with susceptible animals and to avoid contact with other people who deal with susceptible animals (this behavior must be more strict for those people that maintained direct contact with the sick animals);
 - Forbid visits by non-authorized people, including veterinarians, technicians working with artificial insemination and other professionals and producers, particularly those that have contact with susceptible animals;
 - Milk production must be limited to the holding. Do not use these products and by-products to feed susceptible animals (specially calves and pigs). Milk is very important, not only due to the direct risk to the product, but mainly due to the risk of mechanical spreading through trucks and people dealing with milk collection. Irrespective of the produced quantity, exit from the holding must not be authorized because of the risks of spreading the disease. Even knowing that this measure involves many economic and social issues, it should be considered that milk has a very low unit price and, in many cases, it is safer to recommend its destruction, paying indemnity to the producer. Other options to be used and recommended for this product include:
 - Using the milk to manufacture products that undergo thermal processes (mozzarella, cream cheese, among others) on the holding;
 - Milk consumption from healthy animals, after the boiling for at least 5 minutes; and
 - Destruction by chemicals that change the pH (such as vinegar or caustic soda), discarding the product in open pit for this purpose. Do not dispose the product in rivers and other water courses.

Regarding biosecurity, special attention must be given to the disinfectants that will be used in different situations. Appendices 13 and 14, based on PANAF-TOSA Manual of Procedures for the Response to Foot and Mouth Disease and other Vesicular Diseases, details the procedures to be used in cleaning and disinfection in different stages of the veterinary emergency actions and has a summary of the chemicals that must be used for disinfection.

The table below has basic reminders on the main cleaning and disinfection at the entrance and exit of the place where there are suspect cases of infectious vesicular disease:

Basic cleaning and disinfection measures to enter and exit places with a suspect of infectious vesicular disease
<p><u>For professionals team:</u></p> <ul style="list-style-type: none">• At the entrance:<ul style="list-style-type: none">▪ Wear proper clothes▪ Prepare disinfectant solution▪ Wet boots with disinfectant solution• At the exit:<ul style="list-style-type: none">▪ Wash and disinfect boots▪ Undress the clothes used and put them in a plastic bag, as it is done with the material used during the inspection of animals and sample collection▪ Disposable material have to be put in other plastic bags for later destruction
<p><u>For vehicles used:</u></p> <ul style="list-style-type: none">• Clean and disinfect pedals and floor• Wash tires with disinfectant solution• Drive directly to the local veterinary unit

5th Back to the local veterinary unit

After going back to the local veterinary unit, the veterinarian must communicate the result of the investigation to his/her supervisor and other members of the team and fill in the records of activities carried out. Depending on the investigation result, specific actions must be carried out in accordance with the following guidelines:

a) Ruling out vesicular disease

The following actions include: finish filling in the event book, including the diagnosis; file the FORM-IN in a specific file; and submit a copy of the FORM-IN attached to the weekly report to the central unit so that the information is entered on SivCont. It is important to highlight that even false accusations and any other reason for ruling out the case must be included in the weekly report of the zoosanitary information system and on SivCont. The GTA issuing with origin in the suspect holding and in the related holdings for geographic proximity or animal movement must be resumed.

In cases of suspect of other infectious diseases, the investigation must be deepened, including sampling and shipping material for laboratory diagnosis. It is also recommended to do at least one more visit to the holding, in order to evaluate the clinical and epidemiological evolution of the disease event, when a new blood serum sampling (paired sampling) should be taken if necessary. Each visit must be compulsorily recorded in the FORM-COM and the closing of the investigation must be recorded in the completion FORM-COM. The laboratory result must be stored in a specific file, together with the others forms related to the investigated case.

b) Confirmation of the suspect vesicular disease

When a probable case of vesicular disease is confirmed, the ALERT STAGE begins, as described below. Some actions to be immediately carried out in the veterinary unit in charge of the herd with confirmed cases of vesicular disease and by the central unit of the state veterinary service.

At the local office, the veterinarian that received the notification must take the following initial provisions:

- Immediately inform the confirmation of the clinical suspect to the superiors and to the person in charge of veterinary emergencies in the state;

- Maintain the suspension of animal movement from neighboring holdings and holdings related by movement in the last 30 days as of the possible time of onset of the disease;
- Add any supplemental data to the book of events;
- Rewrite the FORM-IN (if necessary) and send it immediately, by FAX or internet, with copy to the central unit of the state veterinary service. The state veterinary service must immediately inform SFA and DSA/SDA/MAPA;
- Prepare and appropriately store the collected material and send it, as soon as possible, followed by a legible copy of the FORM-IN, to the central office of the state veterinary service;
- Make an in depth analysis of animal movements involving the herd with probable cases of vesicular disease. Identify all holdings that, in the last 30 days as of the possible onset of the disease, had relationship due to entrance or exit of susceptible animals with the herd under investigation. The list of holdings in other local veterinary units of the state or other states must be sent to the central unit of the state veterinary service that must make the necessary contacts to carry out a clinical and epidemiological inspection in the herd;
- Define the number of teams necessary to do the clinical and epidemiological inspection on the neighboring holdings or on the holdings near the herd under investigation and communicate the demand to the central unit of the state veterinary service for immediate action. Consider that, in case of FMD, the animals can shed the virus three days before the beginning of the clinical signs, therefore a team for each neighbouring holding must be appointed in order to reduce the risk of the disease spreading. Even if clinical signs compatible with vesicular disease are not observed, under these conditions it is important that all biosecurity procedures are adopted at the entrance and exit of the holdings.

An important subject regarding the management of the activities carried out by the field teams is the risk of the disease being spread by the professionals that inspect the animals. Older studies, such as the one carried out by Sellers and collaborators (1970⁴ and 1971⁵), reported that one person out of eight people exposed to infected animals would carry the virus in their upper airways for over 28 hours and none of them continued to carry it 48 hours after exposure, and that the carriers could infect animals in conditions of not common direct contact in the field (sneezing and coughing on the animals' muzzle). But other two more recent studies show that the use of specific hygiene and biosecurity measures during the disease outbreak reduce the spreading risk by the response teams (Amass and collaborators, 2003⁶ and 2004⁷, Sellers & Gloster, 2008⁸). The authors conclude that long periods are not necessary to prevent the transmission of the FMD virus by people if all organic matter is removed by hand washing, shower and hair washing, cleaning of the upper airways particularly the nasal and tonsil cavities and wearing clean clothes. Therefore, in order to minimize the risks of mechanical transmission as much as possible, and to facilitate the management of the field teams, we recommend that every person who visits a confirmed outbreak or a herd with a high risk of being infected should enter other holdings with susceptible animals only after having taken every biosecurity measure summarized above. In case of intensive handling of animals in a herd with suspect animals, we recommend to visit the other non-infected holding only after 24 hours.

The state veterinary service, after having received the samples and the FORM-IN, must:

- immediately send a copy of the FORM-IN to the corresponding SFA, and to DSA/SDA/MAPA, by FAX or e-mail (dsanimal@agricultura.gov.br);
- SFA and DSA/SDA/MAPA must establish a communication flow to confirm that the FORM-IN was received and to follow-up the developed activities. This flow is coordinated by the Epidemiology Division of DSA/SDA/MAPA, with the support of the FMD Coordination, by telephone and e-mail (dsanimal@agricultura.gov.br) or other electronic system;

⁴ Sellers, R.F., Donaldson, A.I., Herniman, K.A.J., 1970. Inhalation, persistence and dispersal of Foot-and-mouth disease virus by man. *Journal of Hygiene, Cambridge* 68, 565–573.

⁵ Sellers, R.F., Herniman, K.A.J., Mann, J.A., 1971. Transfer of Foot-and-mouth disease virus in the nose of man from infected to non-infected animals. *Veterinary Record* 89, 447–448.

⁶ Amass, S.F., Pacheco, J.M., Mason, P.W., Schneider, J.L., Alvarez, R.M., Clark, L.K., Ragland, D., 2003. Procedures for preventing the transmission of foot-and-mouth disease virus to pigs and sheep by personnel in contact with infected pigs. *Veterinary Record* 153, 137–140.

⁷ Amass, S. P. et al. 2004. Procedures for preventing transmission of foot-and-mouth disease virus (O/TAW/97) by people. *Veterinary Microbiology* 103, 143–149.

⁸ Sellers, R, Gloster, J. 2008. Foot-and-mouth disease: A review of intranasal infection of cattle, sheep and pigs. *The Veterinary Journal* 177, 159–168.

- The state emergency team must be informed and participate in all activities in the alert stage;
- As fast as possible, prepare and send the samples to MAPA's laboratory. The competent department of DSA/SDA/MAPA must keep all state veterinary services informed on the laboratories that carry out the diagnosis of FMD and differential diseases. The state veterinary services must have resources always ready to be used and specific procedures for shipping material to the diagnostic laboratories. The sample shipping to the laboratory must be done after a telephone call informing the means of transportation used and the time foreseen for the arrival of the material.

Appendix 15 describes some recommendations on packaging, storing and shipping infectious material to the laboratory. The samples collected from animals for isolation and identification of pathogen agents causing the diseases described in this document are classified as **UN3373 – BIOLOGICAL SUBSTANCE – category B**, in accordance with the “Guía sobre la regulamentación relativa al transporte de sustancias infecciosas 2007-2008”. In addition, the guidelines and specific requirements defined by the department that coordinates MAPA's laboratory network must be followed. The OIE, through the “Manual de las Pruebas de Diagnóstico y de las Vacunas para los Animales Terrestres” (Chapter 1.1.1), also gives guidelines referred to the collection, storage and shipping of laboratory samples. For further reading, we recommend PANAFTOSA's publication: *Procedimiento para colecta y remisión de muestras para el diagnóstico de enfermedades vesiculares y su diagnóstico diferencial*, available on the PANAFTOSA website under “*Publicaciones del Laboratorio*”.

- Provide the necessary support to the activities related to the alert stage, including the identification of teams for supplementary surveillance and, if necessary, contact the people in charge of other local veterinary units and state veterinary services to inform the identification of the holdings to carry out the inspection.

Figure 5 presents the flow with the stages and activities during the response to a notification of a suspect of vesicular disease. A more detailed copy of this flow must be posted in the central unit, regional units and local veterinary units of the state veterinary services.

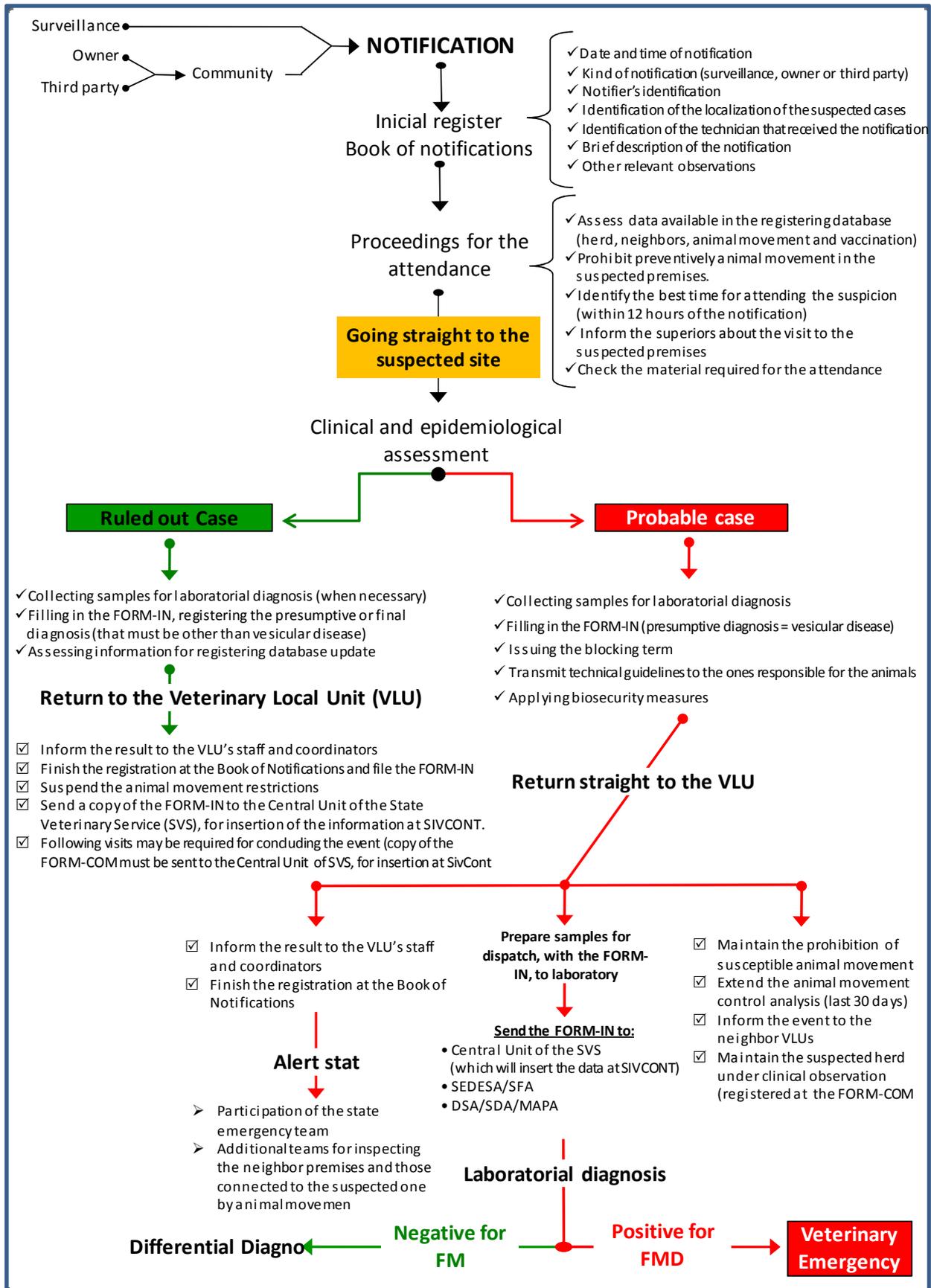


Figure 5. Response flow to the notification of suspects of vesicular diseases

4. Stage 2: Alert

4.1. *Surveillance Activities*

The alert stage comprises the period of time from the confirmation of the probable case of vesicular disease and the final diagnosis confirmed by the laboratory test. As already mentioned, this stage must be carried out considering the real possibility of the occurrence of FMD. Its duration depends on how rapid laboratory diagnosis is, which depends on the quality of the material received. If the involved holding is located on the border of countries or states, the veterinary services of the neighbouring countries or states must be notified. This notification must be made horizontally (between the local veterinary units involved) and intensified by DSA/SDA/MAPA, including notification to PANAF-TOSA when international borders are involved.

It must be carefully done in order not to panic the local population. In this phase of the work, only the professionals who will carry out the preventive actions and supplementary investigation must be involved.

The main purposes are to assess the possibility of the disease occurring in other herds; restrain the movement of susceptible animals to minimize the risks of spreading the viral agent; and continue gathering information to implement veterinary emergency actions if necessary.

The works at this stage must be coordinated by the veterinarian of the local veterinary unit where the herd with probable cases of vesicular diseases is located, with the support of the state team of veterinary emergency. Among his/her responsibilities is the coordination of the support teams to carry out the clinical and epidemiological investigation in other holdings located in their jurisdiction. The priority is to inspect the holdings neighbouring the herd under investigation and the holdings linked due to entrance or exit of animals in the 30 days before the probable onset of the disease. If the owner and other farmers with animals on the holding under investigation raise susceptible animals in other holdings, these holdings are to be identified and included in the surveillance. Once animal's inspection is concluded in the herds considered to be highly risky, the team must wait for 24 hours to inspect other holdings, taking all biosecurity measures described above. In the interval between inspections, the professionals must gather and analyse data.

During inspection, the support teams must adopt all procedures and guidelines in item 3 (investigation stage) of this manual. All activities must be recorded in specific surveillance forms. In herds where cases of vesicular diseases are identified, the staff must fill in the FORM-IN, collect material and draft the Notice of Blocking.

Related holdings with entrance and exit of animals in the last 30 days, involving the herd under investigation, must be inspected and be monitored for at least 14 days.

In case of neighbouring owners, there is a natural tendency to remove the animals from pastures and pens near the holding under investigation, once they are aware of the problem. These producers must be inspected and informed not to do so considering the risks involved in the spreading of the disease to the other animals in their holdings and to other holdings in the region.

In the regions of milk production, special attention must be given to the vehicles that collect milk. Collection lines must be identified and the areas involving the holdings under investigation must be blocked, and alternative routes must be defined.

4.2. *Identification of probable cases of vesicular disease during animal movement*

Although rare, it is a possibility to be considered. In these cases, the procedures depend on different factors such as means of transportation used, quantity of animals involved, place where the problem was identified, distance from the holding of origin; availability of a premise to isolate the animals, etc. Irrespective of how complex the involved factors may be, the following must be considered:

- If animals with clinical signs of vesicular disease were identified in the international border inspection posts, the veterinary service in charge must keep the animals, take samples to send to the diagnostic laboratory and immediately contact SFA, DSA and VIGIAGRO for action regarding the animal health authorities of the country of origin;
- In case the identification of animals with clinical signs of vesicular disease occurred in state border inspection posts, prohibit the entrance in the state, keep the vehicle with the animals and immediately notify the central unit of the state veterinary service. The central office must immediately notify SFA and DSA/SDA/MAPA to communicate the states involved, mainly the state of origin of the animals, in order to perform a joint action;

- The holding of origin of the animals, as well as the neighboring holdings and those related by movement or another epidemiological risk condition, must be inspected and blocked for at least 14 days. Also the holdings located along the path of the animals (mainly if the cattle is walked) must be inspected for at least 14 days;
- Samples for laboratory testing must be taken. For this reason, problems regarding where to take samples and to keep the animals until the laboratory result is disclosed must be solved, considering the following:
 - a) In case of highway movement, the possibility of collecting samples from animals in the vehicle must be evaluated, if safety of professionals and quality of the work carried out is guaranteed;
 - b) Evaluate the possibility of identifying a nearby place for temporary isolation of the animals. This place should not contain other susceptible animals. The choice must consider the risks involved and attempt to find a solution that affects the smallest area possible and that facilitates stamping out if FMD is confirmed;
 - c) if sick animals were found in the holding of origin, and if the holding and the place where movement was prohibited are close, not posing risks to other holdings, the possibility of sending the animals back to their origin should be assessed;
 - d) if the cattle is being walked, the animals must be put in appropriate vehicles to be taken to the area identified for isolation;
 - e) In any case, animals must be transported in sealed trucks escorted by the animal health service and the police. Vehicles must be cleaned and disinfected after the animals are unloaded;
- Material must be collected following all procedures above and the FORM-IN must be filled in.
- In case of confirmation of FMD, the veterinary emergency condition must be established, intensifying investigations in the animals place of origin and pathway taken (remember that, in this case, the problem does not end with the elimination of moving animals, the source of infection has to be identified and eliminated).

4.3. Identification of probable cases of vesicular disease at the slaughterhouse

When there is a suspect of FMD in a slaughterhouse, the veterinarian in charge of the inspection service (federal, state or municipal) has to immediately suspend the slaughter and contact the corresponding local veterinary unit so that samples are collected for laboratory tests. The local veterinary unit has to take the measures described in this document (using FORM-IN to record the actions carried out) and immediately inform the central unit of the state veterinary service that, must immediately inform MAPA through SFA. If there is a Federal Inspection Service (SIF), in addition to the measures described above, SFA has to be immediately informed, and notify DSA and DIPOA/SDA/MAPA.

The measures that must be taken until the final diagnosis of the case are the following:

- Shutdown the plant and stop slaughter;
- Suspend the exit of products from the slaughterhouse, as well as of other materials, objects and vehicles that could vehicle the infectious agent (people movement from the slaughterhouse is subject to the authorization of the official veterinarian in charge of slaughter; and
- Gather information on the origin of the animals and notify the state veterinary service to investigate the involved holdings. GTA issuing will be suspended for movement with origin in these holdings, in holdings related to the suspect holding due to proximity, movement of susceptible animals or another epidemiological risk condition.

Until the disclosure of the diagnosis, with the exception of the batch of animals with clinical signs, the other animals must be slaughtered, and their products and by-products must be kept in the slaughterhouse.

4.4. Identification of probable cases of vesicular disease in livestock events

Once the possibility of vesicular disease in livestock events (fairs, auctions, etc.) is confirmed, the veterinarian in charge must suspend animals receiving and must immediately notify the local veterinary unit and SFA of its state to ensure the implementation of the necessary action specially regarding shutting down the event and sampling material for laboratory tests.

The measures that must be taken until the final diagnosis of the case are the following:

- Restriction to animal movement, vehicles, objects, material and people to prevent the virus spreading;

- Prohibition of the exit of all animals that must remain in their stalls with food and water;
- Sampling to confirm laboratory tests;
- People and vehicles should only exit plant after disinfection and authorization by the veterinarian in charge;
- Suspension of the issuing GTAs to the holdings of origin of the animals with the suspect of vesicular disease, and for those with a relationship due to proximity, movement of susceptible animals or another epidemiological risk condition; and
- Epidemiological investigation in search of the source of infection.

4.5. *Laboratory Result*

The laboratory result may lead to the following situations:

- **Improper material for diagnosis:** due to insufficient quantity or preservation problems. This situation must be prevented, but if it occurs, a new visit to the holding and sampling of material must be immediately arranged (FORM-COM must always be filled in). Information on the new cases of disease must be updated. In the meantime, the holding, as well as the holdings related to it, must remain blocked. In case of old lesions, with difficulty in sampling epithelium, consider the collection of oesophageal-pharyngeal fluid (OPF).
- **Negative diagnosis for FMD and vesicular stomatitis:** in this case, an in depth investigation must be carried out in search of a definitive diagnosis. As it was already mentioned, the capacity of the official veterinary service to reach a final diagnosis is an important element to evaluate the efficiency of the animal health surveillance system. At LANAGRO, the samples submitted will be analysed with other laboratory tests in search of a differential diagnosis. If necessary, there must be a need to go back to the holding and collect new sample for paired analysis. Always remember to fill the FORM-COM during the supplementary visits, updating the information for new cases of the disease. In case the diagnosis of infectious disease is confirmed, keep the lot of animals blocked involved until their complete recovery, giving technical guidance for sanitary control. In this case, a Notice of Blocking Lifting for the holding must be issued. In the suspect of bluetongue, the material with the purpose of isolating the viral agent must be collected. If it is isolated, measures must be taken together with DSA/SDA/MAPA, evaluating the extent of the problem and the activities to be carried out. In this case, the holding must remain blocked. In all cases, the closure of the investigation and the final diagnosis must be recorded in the conclusion FORM-COM, which must be submitted following the same rules of the FORM-IN.
- **Negative for FMD and positive for vesicular stomatitis:** the final diagnosis must consider the isolation of the viral agent. The serological results diagnosis must be based on clinical and epidemiological in-depth analyses, involving the clinical evaluation of a higher number of animals, increasing of the number of samples and investigation in neighbouring holdings.

The disease epidemiology is not completely known. Aspects referred to where and how the virus is maintained in nature and how it is transmitted among the animals and herds are still being investigated. However, considering the current knowledge, once the diagnosis of vesicular stomatitis is confirmed, the following procedures must be carried out:

- Block the holdings with clinical cases for at least 21 days counted as of the healing of the last sick animal (maximum incubation period defined by the OIE). We recommend to isolate animals with clinical signs;
- Considering the possibility of human infection, people that handle sick animals must enhance asepsis and hygiene. Clinical occurrence in humans is rare, but a clinical picture similar to influenza including fever, shivering and muscle pain can occur. The duration ranges from 3 to 4 days;
- Considering the possibility of having insects involved in the transmission of the disease, we recommend to use procedures to reduce the infestation of mainly flies and ticks;
- In milk production areas, biosecurity measures with regard to the milk collection must be adopted to prevent the mechanical spread of the viral agent. In blocked holdings, milk coming from animals with clinical signs must be destroyed. Considering the high sensitivity of the virus to heat treatments, the milk from animals that do not present clinical signs must be removed from the holding and sent to pasteurization or manufacture of products through heat processes;

- Clean and disinfect the area with the highest concentration of animals such as stables, restraining facilities and pens, with the aim of reducing the areas infected by the viral agent. The disinfectants that can be used should contain iodoform, 2% sodium hydroxide solution (caustic soda), 4% sodium carbonate and 1 % formalin solution.
- During blocking, the holdings with clinical cases must be inspected at least once a week, and FORM-COM must be used to record the activities;
- The state veterinary service must carry out an epidemiological investigation including the clinical inspection in herds located near the blocked holdings. If there is a possibility to find other holdings with sick animals, consider the possibility of FMD and use the procedures described in this document;
- By the end of each week, the state veterinary service must write a short summary with the information on the works carried out. The reports must be sent by e-mail to SFA and DSA/SDA/MAPA every subsequent Monday morning;
- After finishing the works, the state veterinary service must draft a final report describing the activities carried out, the structure used and the results obtained, and submit it to SFA and to DSA/SDA/MAPA. Remember that the conclusion must be recorded in the closing FORM-COM, which must be submitted to SFA and DSA/SDA/MAPA following the same rules for submitting the FORM-IN; and
- SFA must appoint at least one veterinarian federal inspector to follow-up the field works.

➤ **Positive for FMD → VETERINARY EMERGENCY.**

In this case, the guidelines and procedures are described in Volume II of this document. The DSA/SDA/MAPA must immediately communicate the occurrence of the disease to the institutions and countries with which it has commercial relationships, to neighbouring countries, particularly to the Permanent Veterinary Committee of the South Cone (CVP), PANAFTOSA, OIE, neighbouring companies and commercial partners. The actions to be taken must depend on the technical, economical and political decisions, particularly considering the region where the outbreak occurred, disease spreading, the available resources for sanitary intervention (chiefly indemnity), the deadlines to internationally regain the status of FMD free (Art. 8.5.8 of the 2008 Terrestrial Animal Health Code) and the possibility of the establishment of a containment zone (Art. 8.5.7 of the 2008 Terrestrial Animal Health Code).

APPENDIX 1 – List of material, equipment and forms for the response and notification of suspect of vesicular diseases.

Template of check list of material available in each local veterinary unit.

List of materials to be used in responses to suspect vesicular diseases	
1 Mouth opener	23 Torches and batteries
2 Antiseptics	24 Surgical and plastic gloves
3 Needles: - Sterilized, disposable hypodermic needles (40 x 16 mm) - Disposable needles for vacutainer® tubes (0.80x25) – include the 25 x 8 mm piece for multiple collection.	25 Overalls
4 Cotton balls	26 Disposable masks
5 Plastic bucket	27 Material for animal identification: ear tags and tagging gun, tattooing needle, etc.
6 Note pad	28 2 ml Eppendorf microtubes (for serum)
7 Backpack sprayer	29 Paper towel
8 Rubber boots (or overshoes)	30 pH strip or <i>Vallée Medium</i> with pH indicator
9 Necropsy instruments	31 Tweezers
10 Styrofoam (thermal) boxes in several sizes	32 Disposable, sterile, 3 ml Pasteur pipette (transfer)
11 Boxes for material transportation	33 Sign identifying that the holding is blocked
12 Disinfectants and detergents	34 Clip board
13 Rubber boots (or overshoes)	35 Box for disposing of needles
14 Brushes for hands and boots	36 Soap/ detergent
15 Sticking plaster/band-aid	37 Plastic bags or bags for waste disposal
16 Polyethylene adhesive tapes	38 Syringe (also syringe for insulin – sampling vesicular fluid)
17 FORM-IN / FORM-COM	39 Sterile swab to collect material for differential diagnosis*
18 Nose ring	40 Clinical thermometers for veterinary use
19 Sterile tubes with <i>Vallée Medium</i>	41 Notice of Blocking and Notice of Blocking Lifting
20 Sterile tubes with EAGLE MEM medium	42 Scissors, scalpel and blades
21 Gauze and bandages	43 10 ml Vacutainer test tubes with and without anticlotting fluid
22 Lasso or ropes for restraining	

*After swabbing, break the tip of the swab and place it in an Eppendorf tube with Eagle MEM medium

Checking Registration		
Date	Name and signature of the professional in charge	Notes
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APPENDIX 2 – Technical disease card on FMD (translation of the OIE text)

Aetiology

Classification of the causative agent

A virus of the family Picornaviridae, genus Aphthovirus
Seven immunologically distinct serotypes: A, O, C, SAT1, SAT2, SAT3, Asia1

Resistance to physical and chemical action:

Temperature: Preserved by refrigeration and freezing and progressively inactivated by temperatures above 50°C.
pH: Inactivated by pH <6.0 or >9.0
Disinfectants: Inactivated by sodium hydroxide (2%), sodium carbonate (4%), and citric acid (0.2%). Resistant to iodophores, quaternary ammonium compounds, hypochlorite and phenol, especially in the presence of organic matter
Survival: Survives in lymph nodes and bone marrow at neutral pH, but destroyed in muscle when is pH <6.0 i.e. after rigor mortis. Can persist in contaminated fodder and the environment for up to 1 month, depending on the temperature and pH conditions.

EPIDEMIOLOGY

- One of the most contagious animal diseases, with important economic losses.
- Low mortality rate in adult animals, but often high mortality in young due to myocarditis.

Hosts

- Bovidae (cattle, zebu, domestic buffaloes, yaks), sheep, goats, swine, all wild ruminants and suidae. Camelidae (camels, dromedaries, llamas, vicunas) have low susceptibility

Transmission

- Direct or indirect contact (droplets)
- Animate vectors (humans, etc.) and Inanimate vectors (vehicles, implements)
- Airborne, especially temperate zones (up to 60 km overland and 300 km by sea)

Sources of virus

- Incubating and clinically affected animals
- Breath, saliva, faeces, and urine; milk and semen (up to 4 days before clinical signs)
- Meat and by-products in which pH has remained above 6.0
- Carriers: particularly cattle and water buffalo; convalescent animals and exposed vaccinates (virus persists in the oropharynx for up to 30 months in cattle or longer in buffalo, 9 months in sheep). African Cape buffalo are the major maintenance host of SAT serotypes

Occurrence

FMD is endemic in parts of Asia, Africa, the Middle East and South America (sporadic outbreaks in free areas)

For detailed information on occurrence, see recent issues of World Animal Health

DIAGNOSIS

Incubation period is 2-14 days

Clinical diagnosis

Cattle

- Pyrexia, anorexia, shivering, reduction in milk production for 2-3 days, then:
 - smacking of the lips, grinding of the teeth, drooling, lameness, stamping or kicking of the feet: caused by vesicles (aphthae) on buccal and nasal mucous membranes and/or between the claws and coronary band
 - after 24 hours: rupture of vesicles leaving erosions
 - vesicles can also occur on the mammary glands
- Recovery generally occurs within 8-15 days.

- Complications: tongue erosions, superinfection of lesions, hoof deformation, mastitis and permanent impairment of milk production, myocarditis, abortion, death of young animals, permanent loss of weight, loss of heat control ('panthers')

Sheep and Goats

- Lesions are less pronounced. Foot lesions may go unrecognised. Lesions in dental pad of sheep. Agalactia in milking sheep and goats is a feature. Death of young stock.

Pigs

- May develop severe foot lesions particularly when housed on concrete. High mortality in piglets a frequent occurrence.

Lesions

- Vesicles or blisters on the tongue, dental pad, gums, cheek, hard and soft palate, lips, nostrils, muzzle, coronary bands, teats, udder, snout of pigs, corium of dewclaws and interdigital spaces.
- Post-mortem lesions on rumen pillars, in the myocardium, particularly of young animals (tiger heart)

Differential diagnosis

Clinically indistinguishable: vesicular stomatitis; Swine vesicular disease; Vesicular exanthema of swine

Other differential diagnosis: Rinderpest; Mucosal disease; Infectious bovine rhinotracheitis; Blue Tongue; Bovine mammillitis; Bovine papular stomatitis and Bovine viral diarrhoea

Laboratory tests

Procedures

Identification of the agent

- ELISA
- Complement Fixation Test
- Virus isolation: inoculation of primary bovine thyroid cells and primary pig, calf and lamb kidney cells; inoculation of BHK-21 and IB-RS-2 cell lines; inoculation of mice

Serological Tests (prescribed tests in the Manual)

- ELISA
- Virus neutralisation test

Samples

- 1 g of tissue from an unruptured or recently ruptured vesicle. Epithelial samples should be placed in a transport medium which maintains a pH of 7.2-7.4 and kept cool (see Manual).
- Oesophageal-pharyngeal fluid collected by means of a Probang cup Probang samples should be frozen to below -40°C immediately after collection)

Note: Special precautions are required when sending perishable suspect FMD material within and between countries. See Manual, Chapter 1.4.

PREVENTION AND CONTROL

Sanitary prophylaxis

- Protection of free zones by border animal movement control and surveillance
- Slaughter of infected, recovered, and FMD-susceptible contact animals
- Disinfection of premises and all infected material (implements, cars, clothes, etc)
- Destruction of cadavers, litter, and susceptible animal products in the infected area
- Quarantine measures (Code Chapter 2.1.1.)

Medical prophylaxis

Inactivated virus vaccine containing an adjuvant. Immunity: 6 months after two initial vaccinations, 1-month apart, depending on the antigenic relationship between vaccine and outbreak strains.

APPENDIX 3 - Foot-and-Mouth Disease

(translation of the text produced by the University of the State of Iowa, USA, updated in June, 2007, available on the OIE website)

Importance

Foot-and-mouth disease (FMD) is a highly contagious viral disease of livestock. It can rapidly spread through a region if control and eradication practices are not implemented upon its detection. Weight loss, poor growth, permanent hoof damage, and chronic mastitis are just some of the sequelae of infection. The detection of FMD in a country impacts international trade and embargoes could cause significant economic losses.

Etiology

The foot-and-mouth disease virus (FMDV) is in the family Picornaviridae, genus Aphthovirus. There are 7 immunologically distinct serotypes and over 60 subtypes. New subtypes occasionally develop spontaneously. The FMDV is inactivated at a pH below 6.5 or above 11. The virus can survive in milk and milk products when regular pasteurization temperatures are used. However, it is inactivated by ultra high-temperature pasteurization procedures. Virus stability increases at lower temperatures and can survive in frozen bone marrow or lymph nodes. The virus can also survive drying and may persist for days to weeks in organic matter under moist and cool temperatures. It is inactivated on dry surfaces and by UV radiation (sunlight).

Species Affected

FMDV primarily affects cloven-hoofed domestic and wild animals, including cattle, pigs, sheep, goats, and water buffalo. Other susceptible species include hedgehogs, armadillos, nutrias, elephants, capybaras, rats, and mice.

Geographic Distribution

Foot-and-mouth disease was found worldwide after World War II. The last U.S. outbreak was in 1929. Endemic areas include parts of Asia, Africa, the Middle East, and parts of South America. Recent outbreaks (2006-2007) have occurred in Argentina, Bolivia, Botswana, Brazil, China, Ecuador, Egypt, Guinea, Israel, Jordan, Kazakhstan, Korea, Lebanon, Palestine, Russia, South Africa and Turkey. In 2001, an outbreak of FMD occurred in the United Kingdom and other countries of Europe; no further outbreaks in the EU have occurred since. North America, Central America, Australia, and New Zealand have been free for many years.

Transmission

Transmission primarily occurs by respiratory aerosols and direct or indirect contact with infected animals. Aerosol transmission requires proper temperature and humidity. The virus may survive for 24 to 48 hours in the human respiratory tract and could serve to spread the virus if precautions are not taken. Animals may also become infected by ingesting animal products contaminated with the virus such as meat, milk, bones, and cheese. Additionally, contaminated objects, such as boots, hands, clothing, vehicles or equipment can spread the virus from animal to animal or farm to farm.

Sheep and goats are considered maintenance hosts. They can have very mild signs; therefore, diagnosis may be delayed, which allows time for spread or environmental contamination. Pigs are considered amplifying hosts, as they can shed large quantities of the virus once infected. Cattle are generally the first species to manifest signs of FMD, so are considered 'indicators' of this disease. Lesions in cattle are typically more severe and progress more rapidly compared to other species.

Ruminants can carry the virus for long periods in their pharyngeal tissue. Recovered or vaccinated cattle exposed to diseased animals can be healthy carriers for 6 to 24 months. Sheep can be carriers for 4 to 6 months. Some strains of the virus can affect one species more than others.

Incubation period

Animals in contact with clinically affected animals will generally develop signs of disease in 3 to 5 days. The virus can enter through damaged oral epithelium or the tonsils in pigs fed contaminated garbage. In this case signs can be seen in 1 to 3 days. Experimental exposure can elicit signs in 12 to 48 hours. Peak time of shedding of the virus and transmission usually occurs when vesicles rupture.

Clinical signs

Foot-and-mouth disease is characterized by fever and vesicles (blisters), which progress to erosions in the mouth, nares, muzzle, feet, or teats. Typical clinical signs include depression, anorexia, excessive salivation, serous nasal discharge, decreased milk production, lameness, and reluctance to move. Abortion may occur in pregnant animals due to high fever (FMDV does not cross the placenta). Death in young animals is due to severe myocardial necrosis. In cattle, oral lesions are common with vesicles on the tongue, dental pad, gums, soft palate, nostrils, or muzzle. Hoof lesions are in the area of the coronary band and interdigital space. In pigs, the hoof lesions are usually severe with vesicles on the coronary band, heel, and interdigital space. Vesicles can also be seen on the snout. Oral lesions

are not as common as in cattle and are usually less severe. Drooling in pigs is rare. Sheep and goats show very mild, if any, signs of fever, oral lesions, and lameness. Animals generally recover in about two weeks with very low mortality in adult animals. Secondary infections may lead to a longer recovery time.

Post Mortem Lesions

The characteristic lesions of foot-and-mouth disease are single or multiple vesicles/bullae from 2 mm to 10 cm in diameter. Early lesions range from a small pale area to a fluid-filled vesicle, sometimes coalescing with adjacent lesions to form bullae. Once vesicles rupture, red, eroded areas or ulcers will be noted. These may be covered with a gray fibrinous coating; a demarcation line of newly developing epithelium may be noted. Loss of vesicular fluid through the epidermis may lead to “dry” lesions. These will appear necrotic instead of vesicular and are more common in the oral cavity of pigs. Lesions also occur at the coronary band and can progress to the skin and hoof. Coronitis may be seen on the hooves and pigs may slough their claws in severe cases. In younger animals, gray or yellow streaking in the myocardium, also called “tiger heart” lesions, may be seen and are caused by zones of degeneration and necrosis in the tissue. Vesicular lesions may also be found on the ruminal pillars.

Morbidity and mortality

In susceptible populations, morbidity can be 100%. Mortality is generally less than 1% but can be up to 40% in younger animals or outbreaks situations.

Clinical Diagnosis

Clinical signs of concurrent salivation and lameness with vesicles and/or erosions should make foot-and-mouth disease a differential consideration. Febrile animals should be carefully examined for early oral or digital lesions. The mouth of any lame animal, and the feet of animals with oral lesions or drooling, should also be checked. Teats of lactating females should be examined. Tranquilization may be necessary for a thorough examination as vesicles may be difficult to see. Laboratory testing is an absolute requirement to confirm FMDV infection as all vesicular diseases have almost identical clinical signs.

Differential diagnosis

The clinical signs of FMD can be similar to vesicular stomatitis, swine vesicular disease, vesicular exanthema of swine, foot rot, traumatic stomatitis induced by poor quality feed, and chemical and thermal burns. In cattle, oral lesions seen later in the progression of FMD (erosions, ulcers) can resemble rinderpest, infectious bovine rhinotracheitis (IBR), bovine viral diarrhoea (BVD), malignant catarrhal fever (MCF), and epizootic hemorrhagic disease. In sheep, these later lesions can resemble bluetongue, contagious ecthyma, and lip and leg ulceration.

Laboratory Tests

FMDV can be identified using enzyme-linked immunosorbent assay (ELISA), complement fixation, and virus isolation. Virus isolation is done by inoculation of primary bovine thyroid cells and primary pig, calf and lamb kidney cells, inoculation of BHK-21 and IBRS-2 cell lines, or inoculation of mice. ELISA and virus neutralization tests can be used to detect antibodies in serum. Virus isolation and identification must be performed on the initial case. Subsequently, antigen or nucleic acid detection can be used to diagnose additional cases in an outbreak.

Samples to collect

Before collecting or sending any samples from vesicular disease suspects, the proper authorities should be contacted. Samples should only be sent under secure conditions and to authorized laboratories to prevent spread of the disease. Since vesicular diseases can not be distinguished clinically, and some are zoonotic, samples should be collected and handled with all appropriate precautions.

Samples include vesicular fluid, the epithelium covering vesicles, oesophageal-pharyngeal fluid, unclotted whole blood collected from febrile animals and faecal and serum samples from infected and non-infected animals.

Recommended actions if FMD is suspected

- Notification of Authorities

A quick response is vitally important in containing an outbreak of FMD. State and federal veterinarians should be immediately informed of any suspected vesicular disease.

- Quarantine and Disinfection

Suspected animals should be quarantined immediately and the premises should be disinfected. Sodium hydroxide (2%), sodium carbonate (4%), citric acid (0.2%), and Virkon-S® are effective disinfectants. Other disinfectants (e.g.,

iodophores, quaternary ammonium compounds, phenols) may be less ideal because they can be rapidly inactivated in the presence of organic matter.

- Vaccination

With seven serotypes, and more than 60 subtypes of FMDV, this task is one of the biggest challenges in FMD vaccination. Currently, there is no universal vaccine against FMD. The U.S., Canada, and Mexico maintain the North American FMD Vaccine Bank, which contains vaccine strains for the most prevalent circulating serotypes in the world. The decision to use vaccination in control and eradication efforts is complex and depends upon scientific, economic, political, and societal factors specific to the outbreak situation. The final decision to use vaccination as an aid in controlling an outbreak of FMD in the U.S., Canada, or Mexico would be made by the Chief Veterinary Officer in each country.

Public Health

FMDV infections in humans are rare, with just over 40 cases diagnosed since 1921. Vesicular lesions can be seen, but the signs are generally mild. Foot-and-mouth disease is not considered to be a public health problem.

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APPENDIX 4 – Technical disease card on vesicular stomatitis (translation of the OIE text)

AETIOLOGY

Classification of the causative agent

Virus family Rhabdoviridae, genus Vesiculovirus
Major serotypes: New Jersey, Indiana

Resistance to physical and chemical action:

Temperature: Inactivated by 58°C for 30 min
pH: Stable between pH 4.0 and 10.0
Chemicals: Ether and other organic solvents sensitive.
Disinfectants: Destroyed by formalin (1%)
Survival: Survives for long periods at low temperatures

EPIDEMIOLOGY

- Morbidity rate variable, up to 90% in a herd
- Low mortality rate

Hosts

- Human (minor zoonosis)
- Domestic hosts: equidae, bovidae, suidae
- Wild hosts: white-tailed deer and numerous species of small mammals in the tropics

Transmission

- Contamination by transcutaneous or transmucosal route
- Arthropod transmission (Phlebotomus, Aedes, etc.)

Seasonal variations: VS is more frequent in the rainy season in tropical areas, although in some countries is also registered during the dry season. Generally disappears at the first frosts in temperate zones

Sources of virus

- Saliva, exudate or epithelium of open vesicles
- Vectors
- Soil and plants (suspected))

Occurrence

The disease is limited to the Americas. (It was described in horses in France in 1915 and 1917, and in South Africa in 1886 and 1887.)

For detailed information on occurrence, see recent issues of World Animal Health, the OIE Bulletin and Weekly and Monthly Epidemiological Report PANAFTOSA/PAHO/WHO

DIAGNOSIS

Incubation period is up to 21 days

Clinical Diagnosis

The symptomatology is similar to that of foot and mouth disease (FMD), with which it can easily be confused (but horses are resistant to FMD and susceptible to VS)

- Excessive salivation
- Blanched raised or broken vesicles of various sizes in the mouth:
 - Horses: upper surface of the tongue, surface of the lips and around nostrils, corners of the mouth and the gums
 - Cattle: tongue, lips, gums, hard palate, and sometimes muzzle and around the nostrils
 - Pigs: snout
- Lesions involving feet of horses and cattle are not exceptional.
- Teat lesions occur in dairy herds
- Foot lesions and lameness are frequent in pigs
- Recovery in around 2 weeks
- Complication: loss of production and mastitis in dairy herds due to secondary infections, lameness in horses

Lesions

Limited to the epithelial tissues of the mouth, teats and feet

Differential diagnosis

Clinically indistinguishable: Foot and mouth disease; Swine vesicular disease and Vesicular exanthema of swine.

Other differential diagnosis: Infectious bovine rhinotracheitis; Bovine viral diarrhoea and bluetongue

Laboratory diagnosis

Identification of the agent

- Virus isolation: inoculation into embryonated chicken eggs; mice; tissue culture systems (chick fibroblasts, pig kidney, BHK-21, Vero); footpad of guinea pigs; horses and cattle; snout of pigs
- Viral antigen detection by complement fixation test, ELISA or neutralisation tests in tissue culture, embryonated chicken eggs, or suckling mice.

Serological Tests (prescribed tests in the Manual)

- Virus neutralisation
- ELISA
- Complement fixation

Samples

Identification of the agent

- Epithelial tissue covering the vesicles placed in buffered glycerol or frozen
- Vesicular fluid aseptically collected and frozen

Serological Tests

- Paired acute and convalescent Serum samples

Note: Serum antibodies reach high levels but reinfection may occur. As for FMD special precautions are required when sending perishable suspect VS material within and between countries. See Manual, Chapter 1.4

PREVENTION AND CONTROL

No specific treatment. Antibiotics may avoid secondary infection of abraded tissues.

Sanitary prophylaxis

Animal movement should be restricted and a laboratory diagnosis must be performed rapidly. Trucks and fomites should be disinfected.

Medical prophylaxis

Inactivated and attenuated virus vaccines have been experimentally tested, but are not yet available commercially.

Note: Differentiation from FMD is very important

APPENDIX 5 – vesicular stomatitis

(translation of the text produced by the University of the State of Iowa, USA, updated in May, 2007, available on the OIE website)

Importance

vesicular stomatitis is an important livestock disease in the Americas. Occasional outbreaks of this zoonotic vesicular disease occur in limited areas of the United States. Affected herds are quarantined until the disease has run its course. vesicular stomatitis closely resembles three foreign animal diseases: foot-and-mouth disease (FMD), swine vesicular disease, and vesicular exanthema of swine. Differentiation of these diseases is important, as a wrong diagnosis could mask the spread of an exotic disease. Prompt diagnosis is also important in containing outbreaks of vesicular stomatitis. The spread of this disease within the U.S. could restrict the exportation of animals and animal products to vesicular stomatitis-free countries.

Aetiology

vesicular stomatitis virus (VSV) is a member of the genus Vesiculovirus in the family Rhabdoviridae. It is a large, bullet-shaped RNA virus. Two strains of VSV are considered endemic to the United States: New Jersey and Indiana-1. Three strains found in South America: Indiana-2 (Cocal), Indiana-3 (Alagoas), and Piry – are considered to be exotic.

Species Affected

Horses, donkeys, mules, cattle, swine, South American camelids, and humans can be affected by VSV. Sheep and goats are relatively resistant and rarely show clinical signs. Experimentally, a wide host range has been found including deer, raccoons, bobcats, and monkeys.

Geographic Distribution

vesicular stomatitis occurs in some of the warmer regions of North, Central and South America, including parts of the United States. Outbreaks also occur occasionally in the more temperate regions of the Western Hemisphere.

Transmission

The transmission of vesicular stomatitis is incompletely understood. VSV is thought to be transmitted by insect vectors, particularly sand flies (*Lutzomyia shannoni*) and blackflies (family Simuliidae). Recently, experimental transmission of VSV (New Jersey serotype) from blackflies to swine was shown to be followed by clinical disease. Transovarial transmission has been demonstrated in both sandflies and blackflies. VSV has also been isolated from mosquitoes. In addition, grasshoppers (*Melanoplus sanguinipes*) can be infected experimentally, and cattle that ingest infected grasshoppers can develop disease. There is also some speculation that VSV could be a plant virus found in pastures, with animals at the end of an epidemiological chain.

Once it has been introduced into a herd, vesicular stomatitis can spread from animal to animal by direct contact. Animals can also be infected by exposure to fomites contaminated with saliva or fluid from ruptured vesicles.

Humans may be infected by contact with the vesicular fluid or saliva from infected animals. Aerosol transmission occurs in laboratories. In addition, some people are probably infected through insect bites.

Incubation period

The incubation period is 2 to 8 days; most often, animals become symptomatic in 3 to 5 days. Occasionally, vesicles can develop within 24 hours. The incubation period in humans is usually 3 to 4 days, but can be as short as 24 hours or as long as 6 days.

Clinical signs

Excessive salivation is often the first symptom. Closer examination may reveal the characteristic lesions – blanched, raised vesicles (blisters) that may be found on the lips, nostrils, hooves or teats, and in the mouth. The vesicle size is highly variable; while some are as small as a pea, others can cover the entire surface of the tongue. A fever usually develops at the time the lesions appear, or just before.

In horses, the vesicles occur most often on the upper surface of the tongue, the gums, the lips, and around the nostrils and corners of the mouth. In some horses, the vesicles may go unnoticed and the disease may appear as crusting scabs on the muzzle, lips, or ventral abdomen. In cattle, the vesicles are usually found on the hard palate, lips and gums, and may extend to the nostrils and muzzle. In both horses and cattle, the hooves may have secondary lesions. In pigs, vesicles usually appear first on the feet, and the first symptom may be lameness. The muzzle is also frequently affected in swine.

Eventually, the vesicles swell and break; the resulting painful ulcers and erosions can cause anorexia, refusal to drink, and lameness. Dairy cattle with lesions on the teats may develop mastitis from secondary infections. Animals can have severe weight loss and, in dairy cows, a drop in milk production. Some cattle may appear to be normal, but eat approximately half of their feed. Unless secondary bacterial infections or other complications develop, the animals recover in approximately two weeks.

Post Mortem Lesions

The necropsy lesions are similar to those in live animals, and may include vesicles, ulcers, erosions, and crusting on the lips, nostrils, hooves, or teats, and in the mouth. Heart and rumen lesions, which may be seen in foot and mouth disease, do not occur in cases of vesicular stomatitis.

Morbidity and mortality

In Central and South America, vesicular stomatitis occurs throughout the year, but it is particularly common at the end of the rainy season. In the south-western U.S, outbreaks of vesicular stomatitis are common during the warmer months, and are often seen along riverways and in valleys.

The morbidity rate is highly variable, and ranges from 5% to 90%. Most cases occur in adults; young cattle and horses under a year of age are uncommonly affected. Deaths are very rare in cattle and horses, but higher mortality rates have been seen in some pigs infected with the New Jersey strain.

Diagnosis

- Clinical

Laboratory diagnosis is essential, as vesicular stomatitis cannot be reliably distinguished from other vesicular diseases including FMD, vesicular exanthema, and swine vesicular disease. However, the presence of symptoms in horses suggests vesicular stomatitis.

- Differential Diagnosis

In cattle, the differential diagnosis includes foot and mouth disease, foot rot, and chemical or thermal burns. The oral lesions can be similar to those of rinderpest, infectious bovine rhinopneumonitis, bovine viral diarrhoea, malignant catarrhal fever, and epizootic hemorrhagic disease. In pigs, FMD, swine vesicular disease, vesicular exanthema of swine, foot rot, and chemical and thermal burns should be considered.

- Laboratory Tests

Detection of the virus or viral antigens is the preferred method of diagnosis. VSV can be isolated in tissue culture, embryonated chicken eggs, or unweaned mice. It can also be isolated by intracerebral inoculation of 3-week old mice. Many cell lines are susceptible to VSV; however, this virus can be differentiated from some other vesicular diseases in African green monkey kidney (Vero), baby hamster kidney (BHK-21) or IB-RS-2 cells. Viral identification in cultures is by immunofluorescence, complement fixation, enzyme-linked immunosorbent assays (ELISAs) and other tests.

In tissue samples, viral antigens can be detected with ELISA, complement fixation or virus neutralization tests. Polymerase chain reaction assays, such as reverse transcriptase–polymerase chain reaction (RT–PCR) may also be used.

The most commonly used serological tests are ELISAs and virus neutralization. Complement fixation, agar gel immunodiffusion, and counter immunoelectrophoresis techniques may also be used.

- Samples to collect

Before collecting or sending any samples from animals with a suspected foreign animal disease, the proper authorities should be contacted. Samples should only be sent under secure conditions and to authorized laboratories to prevent the spread of the disease. vesicular stomatitis is zoonotic; samples should be collected and handled with all appropriate precautions.

Vesicle fluid, the epithelium covering unruptured vesicles, epithelial flaps from freshly ruptured vesicles, or swabs of the ruptured vesicles are the preferred diagnostic samples; APHIS recommends collecting swabs from vesicles. Serum samples, or paired serum samples taken 1-2 weeks apart, may also be collected. In the U.S., paired serum samples are used only for the index case of the nation and index case for each state. Once an outbreak of vesicular stomatitis has been diagnosed in a state, an animal can be declared positive after a single positive complement fixation test.

Recommended actions if vesicular stomatitis is suspected

- **Notification of authorities**

State and federal veterinarians should be immediately informed of any suspected vesicular disease.

Quarantine and disinfection

During an outbreak, state or federal regulations restrict animal movements, and quarantines are placed on facilities with infected animals. Isolation of symptomatic animals helps control the spread of vesicular stomatitis within a herd. If possible, stabling is the preferred means of isolation, as animals on pastures are infected more often with VSV. There should be no movement of animals from an infected property for at least 21 days after all lesions are healed, unless the animals are going directly to slaughter. Insect control may help prevent disease spread. Insect breeding areas should be eliminated or reduced, and insecticide sprays or insecticide-treated ear tags can be used on animals. vesicular stomatitis vaccines are also being tested.

VSV, which is inactivated in sunlight, does not survive for long periods in the environment except in cool, dark places. However, good sanitation and disinfection are necessary to control the spread of the virus on fomites. VSV is susceptible to various disinfectants including 1% sodium hypochlorite, 70% ethanol, 2% glutaraldehyde, 2% sodium carbonate, 4% sodium hydroxide, 2% iodophore disinfectants, formaldehyde and chlorine dioxide. It is also susceptible to UV light, lipid solvents, or heat.

Public Health

Humans may become infected when handling affected animals, contaminated fomites, tissues, blood, and virus cultures. Aerosol transmission occurs, particularly in laboratories. In humans, vesicular stomatitis is an acute illness that resembles influenza. The symptoms may include fever, muscle aches, headache and malaise. Vesicles are rare, but can occasionally be found on the mouth, lips or hands. Deaths have not been reported, and most people recover in 4 to 7 days.

The incidence of vesicular stomatitis in humans is unknown. Although some sources suggest this disease is rare, others point out that human infections may be underreported as they may easily be misdiagnosed as influenza. Approximately 40-46 laboratory-associated infections were documented before 1980, and seroconversion is common.

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APPENDIX 6 – Technical disease card on Swine Vesicular Disease (OIE text)

AETIOLOGY

Classification of the causative agent

Virus family Picornaviridae, genus Enterovirus

Resistance to physical and chemical action

Temperature: Preserved by refrigeration and freezing, inactivated by 56°C/1 hour
pH: Stable over a wide range of pH
Disinfectants: In the presence of organic matter, inactivated by sodium hydroxide (1% combined with detergent). For personal disinfection in the absence of gross organic matter, disinfectants, such as oxidising agents, iodophores, acids etc., are suitable if combined with detergent
Survival: Resistant to fermentation and smoking processes. May remain in hams for 180 days, dried sausages for >1 year, and in processed intestinal casings for >2 years.

EPIDEMIOLOGY

- Morbidity rate in herds may be low but high in groups of pigs (in pens). Does not cause death

Hosts

- Pigs
- Humans: laboratory personnel may seroconvert

Transmission

- Virus readily infects via lesions in skin and mucosa. Direct contact or contact with excretions from infected pigs. Faecal contamination is a major source of virus spread, often within contaminated vehicles.
- Meat scraps and swill derived from infected pigs

Virulent Material

- Intestinal tract is the primary site of infection
- All tissues contain virus during the viraemic period
- Epithelium from vesicles, Vesicular fluid, faeces, and blood of sick animals

Occurrence

The disease has been recorded in Hong Kong, Japan and several European countries.

For detailed information on occurrence, see recent issues of World Animal Health.

DIAGNOSIS

Incubation period is 2-7 days.

Clinical Diagnosis

The clinical signs of SVD may easily be confused with those of Foot and mouth disease (FMD).

- Sudden appearance of lameness in several animals in a group in close contact
- Elevation of body temperature by 2-4°C
- On hard surfaces, animals may be observed to limp, stand with arched back, or refuse to move even in the presence of food. Young animals are more severely affected.
- Vesicles occur on the snout and along the coronary band and interdigital spaces of the feet, and rarely on the epithelium of the buccal cavity, the tongue and the teats.
- Vesicle rupture results in erosions on the skin of the limbs and the coronary bands of the feet. Foot pads may be loosened. Pigs, particularly young stock, may lose the horny hoof.
- Recovery occurs usually within 1 week, with a maximum of 3 weeks.
- Some strains produce only mild clinical signs or are asymptomatic.

Lesions

Vesicle formation is the only known lesion directly attributable to the infection

Differential diagnosis: vesicular stomatitis; vesicular exanthema of swine and foot and mouth disease

Note: Laboratory confirmation is necessary

Laboratory Diagnosis

Identification of the agent: ELISA, direct complement fixation test, cell-culture isolation (pig-derived cell cultures)

Serological tests: Virus neutralisation (prescribed test in the Manual) and ELISA

Samples

Although the virus is very stable, samples must be submitted under the same conditions as those suspected to contain FMD virus, i.e. at pH 7.2-7.4.

Virus isolation

- Vesicular fluid
- Epithelium from vesicles: at least 1 g in PBS containing glycerin 50% (pH 7.2-7.4)
- Unclothed whole blood samples, collected during the febrile period
- Faecal samples from animals with and without lesions

Serological tests

- Serum samples (1-2 ml)
- Also collect serum from other pigs on the premises to test for evidence of subclinical disease

Note: As for FMD, special precautions are required when sending perishable suspect SVD material within and between countries. See Manual, Chapter 1.4.

PREVENTION AND CONTROL

- No treatment
- No vaccination

Sanitary prophylaxis

- Strict quarantine
- Elimination of infected and contact pigs
- Prohibition of feeding with ship or aircraft garbage
- Thorough cooking of garbage
- Control of movement of pigs and vehicles used for transporting pigs
- Thorough disinfection of premises, transport vehicles, and equipment

Medical prophylaxis

Laboratory workers should observe the same caution that applies to any microbiologically contaminated material that may have the potential to cause human infection.

APPENDIX 7 – Cowpox

(text based on documents of the Instituto Mineiro de Agropecuária – IMA)

General aspects

The term “cowpox” is used to describe a contagious disease characterized by the appearance of cutaneous lesions on the udder and teats of lactating cows.

Three different poxviruses can be involved in its aetiology; *cowpox* (real pox), *vaccinia* (the virus used in vaccination against smallpox) and *pseudocowpox* (pseudopox). The lesions are clinically indistinguishable.

It affects cattle of several age groups, lactating cows and suckling calves. Domestic cats and dogs can occasionally be infected. Rodents are considered to be reservoirs.

Transmission between animals is caused mainly by the milker’s hands or by mechanical milking machines. The virus penetrates through lesions on the cows’ teats and udders. It can be transmitted from animals to humans by direct contact with the teat lesions, causing lesions on the milker’s hands and forearms.

The disease is transmitted between holdings by the introduction of infected animals into the herd or by people who milked infected animals on other holdings. Other factors are the handling of infected milk containers and the presence of wild rodents, which can work as virus reservoirs.

The incubation period ranges from 5 to 7 days in humans and animals.

In animals, the occurrence of clinical signs is restricted to lactating cows and calves that suckle from sick cows. The attack rate in lactating cows is usually very high.

In cows, a cutaneous erythema usually develops, followed by the appearance of small spots that become vesicles, and dark crusts on the teats - rarely on the udder - and which heal in 15-20 days. Mastitis and secondary infection in cows are common.

Calves present lesions in their mouths, and on their muzzles and lips. However, lesions are more common on gums and rarely on lips and muzzle.

Humans present ulcerous and pustular lesions, mainly on their hands, but lesions can also occur on the forearms and face, and there can be fever, pain, malaise, and lymphadenopathy.

Among the main problems are the difficulty in milking cows, the decrease in milk production, mastitis, transmission to calves, transmission to humans, and time off work among the milking staff. Regarding economical losses to producers, the most important are the occurrence of mastitis as a consequence of the virus infection, decrease in milk production, the cost of medication, the milking staff being absent for a period of time, making it necessary to hire someone else, and weight loss in calves caused by the lesions after suckling from infected cows.

Diagnosis

Laboratory diagnosis can be carried out with virus isolation, electron microscopy, serology or, in some cases, molecular biology techniques such as Polymerase Chain Reaction (PCR).

To isolate the virus, material obtained from lesions (crusts) is used for inoculation on corioallantoid membrane (CAM) and Vero monolayer cell culture to detect the cytopathic effect. Serum samples collected from infected cows and calves can be submitted to the seroneutralization test. The PCR test for diagnosis of viral agents can be carried out with samples of the total blood, blood serum, milk and faeces.

Crust samples must be collected with tweezers and scissors (gloves must always be worn), and must be placed in an empty sterile flask or on Petri dishes, without adding any other product. **In suspected cowpox, do not sent the lesion epithelium (crusts) in the Vallée Medium**, because this can make the laboratory test difficult. Samples must be collected individually, that is, each flask must contain a sample from one animal only. Each animal’s sample, usually made up of many small fragments, must weigh around 2 grams (the quantity needed to fill a 2 cm square). Animals samples should be collected from untreated animals. If the animal has been treated, state on the FORM-IN the type of treatment carried out on the animal and the products used on the lesions. Immediately after sampling, put flasks or Petri dishes containing samples on ice and then freeze them.

In case of blood serum, it is important to collect samples from animals in the acute stage or in an advanced stage of the disease (healing stage, with crusts). The likelihood of finding protective antibodies in those animals in the advanced stage of the disease is higher. A second serum sampling may be done for those in the acute stage (without crusts on lesions), around 20 to 30 days after the first one. Collect at least 2 ml of blood serum from each

animal. After separation from the coagulate, serum samples must be frozen and sent to the laboratory in Styrofoam boxes with ice.

Treatment, prevention and control

There is no specific treatment for this virus, only support therapy against the symptoms of the disease, that is, the treatment is symptomatic.

No vaccine is available in the market so far. The following procedures stand out among the main prophylaxis and control measures:

- separate infected animals and implement a milking line, in which infected animals are milked and handled last;
- use rubber gloves with enhanced grip patches to dairy cows;
- between cows (i.e. after milking each cow) clean and disinfect the milker's hands and gloves, with a 5,000 ppm chlorine solution (for example 1 liter of bleach mixed with 3 liters of water), in the following process: wash hands with water and soap, put them in the bleach solution and, lastly, rinse with clean water;
- use glycerin iodine on the udder lesions, do not let the calf suckle for at least 2 hours after the product has been applied. Do not use a corticoid ointment;
- tell people who have been infected to go to a local health station;
- contact the municipal health service and inform them of the occurrence of the disease in the region; and
- stop the movement of lactating cows and sucking calves until the complete recovery of the whole herd, preventing the spread of the disease to other holdings through the exit of sick animals.

Milk contamination is neither clear nor proven. Disease transmission by ingestion is unknown. Milk may therefore be sold. However, to prevent risks, consumption is only recommended of boiled or pasteurized milk. Containers used to store and transport milk must be cleaned and disinfected with a bleach solution (particularly the handles).

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APPENDIX 8 – Basic guidelines for examining animals with suspected vesicular disease

1. Restrain the animals suitably and record every detail on signs and lesions observed
2. For every kind of susceptible animal
 - a) Before immobilizing the animals, observe
 - Apathy
 - Signs of lameness
 - Drooling
 - Smacking of the lips
 - Grinding of the teeth
 - b) Record the body temperature and the estimated age

Normal value (possible variation of + or – 0.5°C)

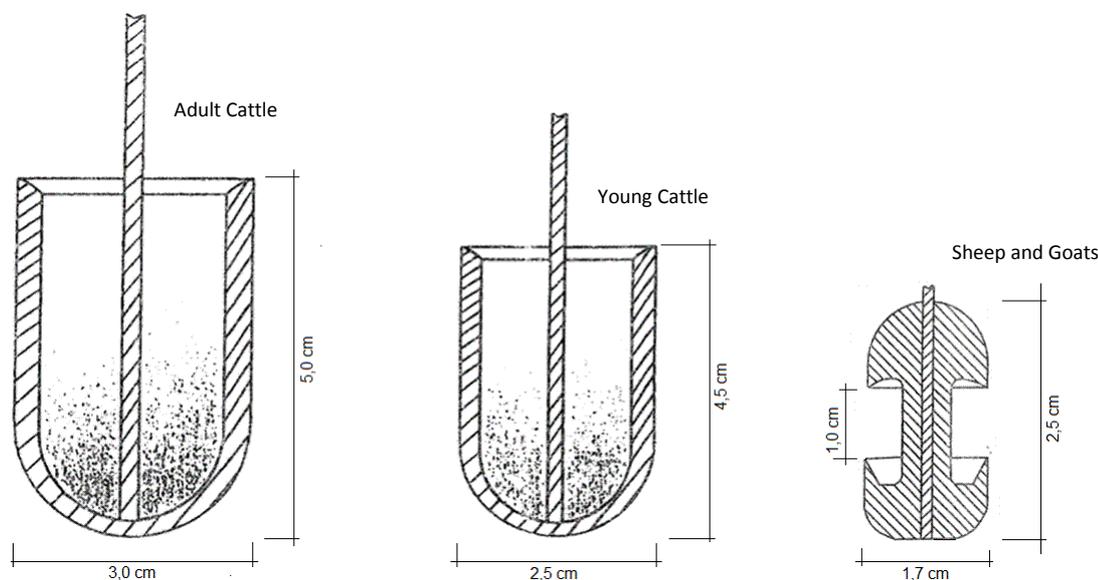
 - Cattle = 38.5°C
 - Sheep = 39.5°C
 - Goats, pigs and horses = 39.0°C
 - c) Describe the vesicles in detail
 - Ruptured or unruptured (open or closed)
 - Size
 - Color (i.e.: whitish, bright red, yellow etc.)
 - Depth
 - Borders (edges) well-defined or poorly differentiated
 - Degree of healing (presence of fibrin deposit)
3. Cattle
 - Clinical History
 - Ask owner about the onset of clinical signs
 - Ask owner about reduction in milk production
 - Location of lesions
 - Inspect nostrils
 - In the buccal cavity, inspect tongue, lips, gums and lateral and upper walls
 - Feet (if necessary, wash with running water): interdigital space, coronary band and heels
 - Udder and Teats
 - Vulva and prepuce
4. Pigs
 - Important signs
 - Acute and sudden lameness
 - Observe the animal on concrete or another hard surface and make it walk
 - Lesions
 - Snout, lips, tongue (lesions are usually smaller and less visible than in cattle), feet (there can be claw detachment from the coronary band)
5. Small ruminants
 - Important signs
 - Acute and sudden lameness (it usually affects every limb), differential diagnosis: *foot-rot*
 - Lesions
 - Usually on the feet, coronary band, but also on the interdigital space and claw loosening
 - Small vesicles at the base of the teeth and on lips
6. Record the data on breed, sex, age, breeding system etc
7. Record animal identification correctly. Remember that every animal sampled must present a permanent or long-lasting individual identification
8. Record all information legibly and check the quality and accuracy of the text

APPENDIX 9 – Technique and procedure for collection of oesophageal-pharyngeal fluid (OPF)
(Text extracted from the publication of Ivo Gomes and Paulo Augé de Mello, February, 1997, mimeographed)

Plants

Plants must be suitably designed for perfect restraining of animals, enabling the head to be immobilized, looking upwards, maintaining an appropriate and comfortable position for collection. The correct restraining of animals is an important factor to facilitate the work and prevent accidents both for animals and for operator.

Oesophageal-pharyngeal fluid (OPF) samples must be collected with the help of specific collectors^{9,10}, as shown in the Figures below. Probang collectors are round-bottomed rounded-rim stainless steel cups fixed to the end of a curved rod of around 50 cm in length.



Types of Oesophageal-pharyngeal fluid (OPF) Cups

Animals

eliminate food remnants and to moisten the oesophageal-pharyngeal region. This will facilitate insertion of the cup and abrasion of the mucosa. Avoid the use of tranquilizers causing muscle relaxation. The animal may retch when the cup is inserted, impairing sample collection. In this case, the operator must reject the material and try to collect again after resting the animal for some hours. If this continues, we recommend postponing the collection to another day.

Sample collection

During work, the operator must take precautions to prevent transmission of the virus from animal to animal such as asepsis, overalls and sterilized boots. Use one sterilized cup for each animal. In order to introduce the cup, the operator has to open the animal's mouth, pressing down on the tongue, and carefully insert the cup through the labial commissure until it reaches the pharynx and the front of the oesophagus. Once the cup has been introduced, it is necessary to make a gentle abrasion of the esophageal-pharyngeal mucosa (three to four times) before removing it. This procedure is critical for sample collection, because the main sites of FMD replication are on the anterior floor of the pharynx and on the dorsal surface of the soft palate¹¹. In a positive animal, the FMD virus must be present in the epithelial cells that detach from the oesophageal-pharyngeal region at the moment of abrasion, together with the presence of the saliva, mucus and food remnants. After collection, the operator must wash his/her hands and arms with a disinfectant solution then rinse with running water.

Conservation and shipping of samples

After removing the cup, its contents have to be transferred to a sterilized wide-mouthed, screw cap 30 ml flask, then immediately add the same quantity of Earle's medium, containing 2x the concentration of antibiotics and fungicide. The flask is identified and duly closed with adhesive tape, and then vigorously shaken to thoroughly blend the sample and the medium. The sample is then placed in a refrigerated container with ice and salt, or dry ice. In this case, make sure that the flask is very well closed to prevent acidification of the sample by carbon dioxide. Samples must be sent as soon as possible to the laboratory, with a protocol identifying the animals and stating the times when collection began and ended. Once received at the laboratory, if not processed immediately, they must be kept in -70°C freezer or -196°C liquid nitrogen, to be preserved until their use.

⁹ Bekkum van *et al.* (1959) Observations on the carrier state of cattle exposed to foot-and-mouth disease virus. *Tijdschrift Diergenesh.*, 84:1159-1164.

¹⁰ Burrows, R. (1968) The persistence of foot-and-mouth disease in sheep. *J. Hyg., Camb.*, 66:633-640.

¹¹ Burrows, R. (1966) Studies on the carrier state of cattle exposed to foot-and-mouth disease virus. *J. Hyg., Camb.*, 64:81-90.

APPENDIX 10 – Composition of solutions used to preserve the material to be sent to laboratories
(source: PANAFTOSA)

Vallée Medium 50%

(for epithelium sampling)

1. KH_2PO_4 (1.35g)
 2. K_2HPO_4 (7.80g)
 3. H_2O demineralized – c.s.p. (1,000ml)
 4. Glycerol (mix at the end) (1,000ml)
 5. Phenol red (for pH control)
- Autoclave separately for 20 min. at 121 °C the Glycerol and the Phosphate buffer. Mix after autoclaving.
 - pH 7.6
 - Store at 4°C

Earle Medium

With Lactoalbumin Hidrolyzate and Yeast Extract
(for collection of Oesophageal-pharyngeal fluid)

1. NaCl - 80.0g
2. KCl - 4.0g
3. $\text{Mg SO}_4 7\text{H}_2\text{O}$ - 2.0g
4. $\text{NaH}_2\text{PO}_4 \text{H}_2\text{O}$ - 1.4g
5. GLUCOSE ANHIDROUS - 10.0g
6. LACTOALBUMIN HIDROLYZATE (dissolve separately) - 50.0g
7. YEAST EXTRACT - 10.0g
8. NaHCO_3 - 22.0g
9. CaCl_2 (Dissolve separately and add at the very end) - 2.0g
10. Phenol red solution 1%- 10.0ml
11. H_2O desmineralized c.s.p. – 10,000ml

- Adjust final pH to 7.4 - 7.6 with NaOH or HCl/1N
- Filter through a 0.22 μm membrane

OBS.: Double the amount of normal concentration antibiotics at the time of use

EAGLE MEM medium

(for sampling of material for IBR and DVB)

1. MEM powder (GIBCO-61 100) - 9.5 g
2. Sodium Piruvate - 0.11 g
3. Sodium Bicarbonate - 1.5 g
4. Non-essential Aminoacids solution - 10 ml
5. Neomycine Sulfate - 0.22 g
6. Peniciline G Potasic – 100,000 U.I.
7. Fungizone - 2.5 mg
8. Bovine fetal serum - 100 ml
9. H_2O desmineralized c.s.p. - 1.000 ml

- Adjust final pH to 7.5
- Filter with a 0.22 μm membrane

OBS.: Double the amount of normal concentration antibiotics at the time of use

APPENDIX 11 – Template of Notice of Blocking

Space for identification of the state veterinary service

NOTICE OF BLOCKING

Blocking start:

Time:	Day/month/year:
-------	-----------------

 Control Number:

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Place and identification of the blocked plant:

State	Name of the city:	
Name of the plant:		Registration code of the state veterinary service* :

*** State Veterinary Service**

Owner's name:

--

Representative's name:

--

Reason for blocking:

Suspected occurrence of: _____

Occurrence of: _____

Other reason: _____

Legal basis for Blocking: _____

The animal owner, or his/her representative, has been notified that, until further notice, by decision of the official veterinary service:

- FMD susceptible animals must not enter or leave the establishment (cattle, buffalo, sheep, goats and pigs);
- Removal of animal products or by-products, as well as any movement of non-susceptible animals originating on the holding must occur through a specific authorization; and
- There must be no contact with other animals susceptible to FMD for at least 72 hours, avoiding visits to other holdings during this period (guidance valid for every person on the holding who has had contact with infected animals)

Failure to comply with this ban will lead to legal penalties for the offender.

This notice was drafted in 2 (two) copies of the same content, and is signed by the veterinarian, the owner, or his/her representatives, and witnesses.

Signature of the owner or his/her representative:

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Name and signature of the veterinarian of the State Veterinary Service:

--

Witnesses:

Name:	Signature
Name:	Signature

Place and date _____



APPENDIX 12 – Template of the Notice of Blocking Lifting

NOTICE OF BLOCKING LIFTING

On this date _____ in the city of _____,
(Symbol of the State), on the holding named _____
_____, belonging to _____
_____ the veterinarian of the official
service identified below appeared and issued this Agreement, ending the effects of the Notice of Blocking nº
_____ issued on ____/____/_____.

Identification and signature of the veterinarian of the official service

APPENDIX 13 – Summary of disinfectants for FMD

(Adapted from the “*Manual of Procedures for the Response to Foot and Mouth Disease and other Vesicular Diseases*”, PANAFTOSA)

1. 2% Citric acid

Preparation: 2 parts of citric acid to 98 parts of water.

Indications: Laboratory ware and vehicle cabins.

Observation: It is not very corrosive for metals and painted surfaces.

2. 4% Sodium carbonate Solution

Preparation: Dissolve 400 g of sodium carbonate in 10 litres of water.

Contact Time: 10 minutes.

Application Method: spraying, sprinkling, foot bath and immersion.

Precaution: When applying the disinfectant in closed environments, use boots, gloves and mask.

Limitation: acts only in powder.

Indications: plants, people and animals, vehicles, clothing, tools, leather, skin, bones, hay and straw.

3. 10% Formaldehyde solution

Preparation: Dissolve ½ litre of commercial formalin (40% commercial formaldehyde solution) in 5 litres of water.

Contact Time: 30 minutes to 3 hours

Application Method: spraying, sprinkling and immersion.

Indications: clothing, leather, bone, hay and straw.

Precaution: Wear a mask. When using the formaldehyde gas to fumigate a room or a building, the premise must be reasonably airtight. 500 g of potassium permanganate and 0.5 litres of formaldehyde (40% formaldehyde solution) are necessary for every 30 m³ of space. Put the permanganate in an open container (i.e. a can) and add formaldehyde right before closing the premise. Do not put more than 1 litre of formaldehyde in each container. The container must be made of metal (not of glass or plastic, because it generates a lot of heat) and which must be put inside another larger container, also made of metal. The gas has to act as long as possible and never for less than 10 hours. The reaction can produce combustion. The external metallic container has to be three times higher than the internal one and has to be at a distance bigger than 0.50 m from any inflammable metal. On wooden floors, containers have to be put on a protection of tile or metal. Put Hazard Warnings on the doors of the premises.

4. 2% sodium hydroxide solution (caustic soda)

Preparation: Dissolve 200 g of sodium hydroxide in 10 litres of water.

Contact Time: 30 minutes.

Application Method: sprinkling.

Precaution: use boots and gloves.

Limitation: very corrosive.

Indications: plants, manure pits and fences.

5. Iodophor Composition.

Preparation: Mix 1 litre of the product in 200 litres of water.

Contact Time: 10 minutes.

Application Method: spraying, sprinkling, foot bath and immersion.

Indications: People, animals, vehicles, clothing, tools, leather, skin, bone, hay, straw and manure pits.

We have to point out that in animal health emergencies to eliminate FMD outbreaks in Brazil between 1997 and 2005, the products chosen for several purposes were iodophor-based. These products can be easily purchased, preserved and used, and can be used as both disinfectants and antiseptics only by changing the concentration/dilution according to the manufacturer’s recommendations. More recent products also contain detergents that increase the penetration power of the chemical agent, even in the presence of organic matter and substances that reduce the typical corrosive action of most disinfectants available in the market. If detergents with the composition of this disinfectant are not available, an alternative to improve its action with organic matter is to mix 1 litre of domestic detergent to 10 litres of disinfectant.

6. 2% Acetic Acid

Preparation: 2 parts of acetic acid glacial to 98 parts of water.

Indications: Laboratory ware and vehicle cabins.

Observation: not very corrosive for metal objects, but little action on organic matter.

7. 4% Metasilicate

Preparation: 4 parts of metasilicate to 96 parts of water.

Indications: It acts in the denaturation of the protein and its oxidising activity is less than one of a comparable concentration of sodium hydroxide. It is not as corrosive or irritating as sodium hydroxide. It is usually used combined with other disinfectants.

8. 5% Calcium oxide solution 5% (burnt lime)

Preparation: Dissolve 500 g of calcium oxide in 10 litres of water.

Contact Time: 6 to 24 hours.

Application Method: sprinkling, whitewash.

Precaution: use boots and gloves.

Limitation: it is recommended to use it immediately after preparation.

Indications: plants, vehicles, manure pits, walls and pillars. Application recommended after burying animals, on top of the trench and never inside it.

9. 10% commercial creolin solution

Preparation: Mix 9 litres of water with 1 litre of 10% commercial creolin.

Contact Time: 2 hours

Application Method: spraying, sprinkling.

Indications: plants, vehicles and manure pits.

10. Triple salt of potassium monopersulfate solution

Preparation: dilute the powder in running water, 1 part to 1,300 parts for FMD virus.

Contact Time: 30 minutes.

Application Method: spraying, sprinkling (droplets) and immersion.

Precaution: it is not toxic nor irritating.

Indications: Disinfection of stables, pens, industrial processing plants, animal limbs and feet, vehicles and farm equipment.

Limitations to use: Do not mix with alkaline substances, because the product works at 2.5 pH for a solution at 1%. Once prepared, the solution is active for around 7 days.

Notes:

- Effectiveness of citric acid or sodium carbonate solutions improves by adding a small quantity of an appropriate detergent. Add one tablespoon of homemade liquid detergent to 5 litres of water for washing. You can also add one and a half teaspoons of a non-ionic detergent to 10 litres of citric acid solution. The viricidal action of acid or alkaline disinfectants depends on the concentration of the hydrogen ion (pH) in the recommended aqueous dilution. Citric acid and sodium carbonate solutions must have pH <4 and >10 respectively when prepared as above.
- A simple method to establish the concentration of hydrogen ions is to measure the pH with litmus paper. Wet a piece of litmus paper in the disinfectant and put it on a white non-absorbing surface. After 30 seconds, compare its colour to a scale in the package. This pH verification has to be made at random during disinfection. Employees must keep four sets of litmus paper (two for 2-4 pH scale and two for 8-10 pH scale).
- Because the viricidal action of acid or alkaline disinfectants depends on their pH, it is important to keep them separate. Surfaces treated with one type cannot be submitted to the action of another type, unless washed with water in the interval. Never use washing soda and an acid to disinfect the same article.
- Disinfectants recommended for FMD are not effective for many bacteria and pathogen viruses and lose their specific effectiveness if mixed or applied with regular disinfectants.

APPENDIX 14 – Disinfection procedures in FMD cases

(Adapted from the “*Manual of Procedures for the Response to Foot and Mouth Disease and other Vesicular Diseases*”, PANAFTOSA)

It is not possible to lay down definitive rules to cover every point that may arise during an outbreak as far as disinfection is concerned, and common sense should be used in dealing with all problems that might occur.

Disinfection procedures depend on a variety of circumstances in each case, such as the structure of stables or sties, places that infected or suspected animals have access to, the quantity of manure and other impurities, the nature of products considered infected etc.

The most important factor to ensure the inactivation of a causal agent in an infected holding is cleanliness and thorough washing before applying a disinfectant.

Bear in mind that almost every substance used in the disinfection is toxic to a greater or lesser degree. Therefore, institutions involved must take appropriate action to protect the health of workers involved in disinfection.

Gloves, boots and special clothing as well as masks against gases should be worn when working with substances producing fumes. At the end of the task, wash hands, face and exposed surfaces with water and soap. The clothes used in the task must be changed. It is important to keep a first-aid case along with the disinfection equipment, in which there must always be products such as boric acid, carbolic acid, ointments and creams against burns, as well as gauze, cotton wool, iodine etc.

Another precaution to take into consideration concerns the *modus operandi*. Disinfection has to be always carried out upwind, in other words the operator has to take up a position in such a way as to let the air circulate forwards from behind his/her back, avoiding the solutions used in the disinfection being blown toward him by the wind.

Here are some specific recommendations for certain objects and premises to be cleaned and disinfected:

- Livestock buildings and plants

As a preliminary measure before removing manure or another material from the building or from the plants, their contents as well as surrounding land must be wetted with an approved disinfectant.

All parts of buildings and plants that may have come into contact with animals or their excretions must be well scraped and brushed.

Manure, loose litter, fodder etc. must be removed and, if in small quantities, have to be buried or piled up, after being wetted with a disinfectant. If there is a large quantity, it must be gathered in one place where people or animals do not have access and its surface must be thoroughly wetted to prevent wind dispersion. If this is not possible, take it to the nearest convenient arable land where it can be buried immediately. However, public routes cannot be taken for this purpose. Another possibility, if animals are slaughtered and buried on the holding, is to keep a space in the animal trench to bury the material removed from pens and other places.

When the flooring of the buildings is made of earth, clay or chalk, or is permeable to water, the surface has to be removed and carefully wetted with a disinfectant.

Any wooden structure capable of retaining virulent material and that does not allow sufficiently effective disinfection must be removed and burnt. If wooden floors cannot be disinfected, remove and burn them, and turn over the subsoil to a depth of more or less 25 to 30 cm and mix it with lime.

Check whether the disease can be spread by water courses that flow through or into fields holding susceptible animals. Drainage pipes must be closed while there is the risk of spreading the disease and the material has to be disinfected before its extraction.

Animal excrement or fluids from drainage of stables, pens and other places holding susceptible animals must be mixed with sodium carbonate, until obtaining a solution of approximately 4%. This mixture has to be shaken and removed after at least 5 hours.

The owner must be notified in writing of the destruction of objects or parts of the holding, and the amount of compensation agreed upon. Avoid all unnecessary destruction.

Insects and rodents can be used as mechanical vectors. At the beginning of the cleaning and disinfection, rodents move to other buildings. Carry out a prior check to determine the need to control insects and rodents.

- Pens and other places

Walls, fences etc, have to be washed with a disinfectant at the beginning and then scraped and brushed and then washed again. Manure surfaces have to be wetted with the disinfectant recommended for the purpose.

If the manure covering is thick enough to heat up without being piled up, it can remain in situ. Otherwise, it must be removed, starting from the edges to the middle of the yard and then heaped up in order to be then covered with a 4% sodium carbonate solution.

As far as may be reasonably practical, disinfect with 4% sodium carbonate solution in feeding troughs, gates, fields and so on through which infected animals may pass.

Every area that may have come into contact with slaughtering has to be carefully covered with 4% sodium carbonate solution.

- Hay and bales of straw

Surfaces possibly exposed to contamination must be removed and destroyed. The remainder must be mixed with 10% formaldehyde solution.

Where large quantities of forage are suspected of being contaminated, and because they are hard to wash or fumigate, the choice is between destruction, storage for a safe period of time or direct removal to a manufacturing plant. Special attention must be given to hay stored in the upper stories of stables.

- Tubers

Where there is a risk of contamination, deposits where tubers are kept and surrounding floors must receive a 10% formaldehyde solution and, if these deposits are open, exposed tubers must also be disinfected. Tubers from infected places must be cleansed and mixed with 10% formaldehyde solution. Tubers that may have been in contact with infected animals must not leave the holding.

- Other food products

In accordance with their quantity and type, and the possibility of contamination, mix or fumigate them with formaldehyde. Small quantities of food may be discarded, washing it and feeding it to non-susceptible animals on the holdings (birds, horses).

- Bones

Bones on infected holdings and with commercial purposes have to be disinfected, washing them with 10% formaldehyde solution or, if convenient, by fumigation with formaldehyde, and directly sent to companies on closed and sealed trucks.

- Leather and skin

Leather and skin may be removed from infected holdings if steeped in a hot solution of 4% sodium carbonate for 15 minutes or into a 1 x 10,000 sodium bifluoride for 24 hours.

- Working Animals

Horses may continue working on infected holdings, or if necessary, can leave them after previous washing and disinfection of their hooves.

- Milk containers in the infected area

The method which is currently used in milk plants and warehouses to sterilize containers consists of turning them upside down and submitting them to a steam jet for one minute. Their covers are immersed in boiling water for the same time. However, in this system, the temperatures that are reached on the outer surface and at the bottom of the containers are not enough to destroy the FMD virus. Owners and managers of milk plants or milk collecting establishments are therefore recommended to sterilize their containers by immersion in boiling water or to submit the interior to steam and to disinfect the external part. The best sterilization method is immersion in boiling water tanks.

- Contaminated Wool

Disinfection with 2.5% formaldehyde solution for 1 hour at 38 and 40°C, or for 3 hours at temperatures between 18 and 20°C.

- Transport vehicles

Wash the coachwork with disinfectant; carefully scrape and brush off the manure and adhered dirt with special care for edges and angles. Wash the whole structure again with disinfectant. Vehicle wheels must be carefully disinfected.

- Boats and airplanes

A 4% sodium carbonate solution with 0.05% sodium silicate is recommended.

APPENDIX 15 – Requisites for packages, preservation and samples shipping for laboratory tests

Packages have to be of a good quality, strong to resist to loads and impact during transportation, including overflow, piling up, manual or mechanical handling. Packages have to be built and closed to prevent any content loss in normal conditions of transportation by vibration or temperature, humidity or pressure change.

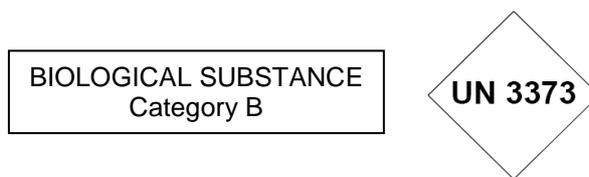
Apply the package system of triple bottling, including for local transportation on surface including three elements: a primary container, a secondary package and an external, obligatory rigid package.

The primary container has to be packed on an absorbing material to contain the whole content without committing the integrity of the damping material nor the secondary package. The primary container has to be protected by a secondary package that, in normal conditions of transportation, cannot rupture or perforate. If Various fragile primary containers are inserted in the same secondary package, they have to be individually packed or separated in such a way to avoid contact between them.

Always use plastic or glass flasks of good quality with screw covering. Serum has to be preferably inserted in rejecting plastic micro-tubes of type *ependorf* of 1.5ml. Be careful to fill just 2/3 of its capacity, because, once frozen the fluids expand its volume.

The secondary package has to avoid the content loss in the case of an error in the primary container and will be inserted in an external package with an appropriate damping material. A copy of the sample list has to be fixed on the secondary package that has to have also the FORM-IN or FORM-COM containing the relevant epidemiologic information of the case, in order to allow the association and identification of the respective samples consigned.

For transportation, the brand “UN3373” (Picture below) has to be fixed on the external surface of the third package, and it shall be easy to be seen and read. The brand shall have a lozenge shape, with a side longer or equal to 50 mm, and lines thickness of at least 2 mm and letters and numbers height of at least 6 mm. The official designation of transportation “Biological Substance, category B”, in letters of at least 6 mm, shall be place on the side of the lozenge brand.



At least one of the external package surfaces shall have the minimum size of 10cm x 10cm.

Primary containers shall not contain more than 1 liter, as well as the secondary package has to be waterproof and drained. The external package shall not contain more than 4 liters or 4 kg, excluding the wet ice, the dry ice or liquid nitrogen when used to conserve samples.

The secondary packaging must withstand pressure of up to 95 kPa (0.95 bar), without spilling.

Ice or dry ice (carbon dioxide) when used must be placed outside the secondary packaging, in other words inside the external package or inside an shipping container (Styrofoam box). Place internal wedges to keep packages immobilized when the ice melts or evaporates. With regular ice, the outer packaging and the shipping container must be waterproof and watertight. With dry ice, the packaging must allow the exit of the gas and prevent an accumulation of pressure that could cause it to burst and must be identified as “Solid carbon dioxide” or “Dry ice”.

When liquid nitrogen is used as coolant, the outer packaging must bear a label identifying the corresponding danger and, when transported by air, the handling label for cryogenic fluids must also be attached.

With an outer shipping container, labels appropriate for external package have to be attached to the shipping container. It also has to be identified with the term “shipping container”.

The outer packaging must also bear a label with the name, address and telephone of the addresser and addressee.

Transportation of samples that are unlikely to contain infectious substances, such as serum and blood for seroepidemiological investigation, or in which pathogenic agents have been neutralized or inactivated to prevent health risks, need not comply with this regulation; they need only assure that the primary packaging is watertight and waterproof, the secondary packaging can be a hermetically sealed plastic bag and the outer label need only bear the sentence “Exempt Animal Sample”.

Training and awareness is important for all personnel involved in the transportation of category B biological substances. Only through appropriate guidance and training can senders ensure the correct classification of the substance to be shipped, as well as the correct choice and preparation of the package. Transporters and other companies employing workers who intervene in this shipping process must train their employees in the appropriate procedures to recognize and handle packages containing biological samples and how to deal with spills, protecting themselves against exposure.



MINISTRY OF AGRICULTURE, LIVESTOCK AND FOOD SUPPLY
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