SEACFMD

A Field Manual for
Animal Disease Outbreak Investigation
and Management

Food and Agriculture Organization of the United Nations

OIE WORLD ORGANISATION FOR ANIMAL HEALTH

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1. Introduction to outbreak investigation and management

This manual on outbreak investigation is intended as a guide to assist field veterinarians and veterinary para-professionals to investigate and manage infectious disease outbreaks in livestock. The manual also provides a reference for those developing contingency plans for infectious disease outbreak management.

The manual presents a step-by-step guide to investigating a disease outbreak. While the methods can be applied to any outbreak situation, this manual focuses on transboundary diseases, particularly foot-and-mouth disease (FMD) and classical swine fever (CSF), given the importance of these diseases in South-East Asia.

The step-by-step guide is supported by background information on why outbreaks occur, the epidemiology and clinical signs of FMD and CSF, reporting procedures, sample collection and submission, and control measures. Generic checklists and forms are provided in the appendices of the manual, which can be copied and adapted for use in animal disease preparedness, outbreak investigation and management scenarios.

1.1 The importance of outbreak investigation

The primary reason for conducting an outbreak investigation is to identify the source of a disease, in order to guide control measures and limit disease spread. Information gathered may also be used to evaluate existing prevention strategies and identify ways to prevent future outbreaks. Outbreak investigations provide an opportunity to document three key pieces of information:

– How quickly the disease is spreading within a population of interest;

– The spatial distribution of disease-positive and disease-negative enterprises; and

– Characteristics or behaviours that are more common among the disease-positive enterprises, compared with those that are disease-negative.

These three key pieces of information allow adaptation of standard disease control procedures to best suit the particular situation and environment, thus resulting in more effective disease control measures.

> **Why investigate an outbreak?**
> – To identify the source(s) of infection.
> – To prevent further exposure of animals to the infection source(s).
> – To determine where the disease has spread.
> – To guide control measures during and after the outbreak.
> – To improve knowledge on the behaviour and pattern of disease.
> – To improve knowledge of the risks and routes of introduction.
> – To provide information that will allow stakeholders to better understand why outbreaks occur in their area, and factors that influence the severity of outbreaks if and when they occur.

1.2 How long should the investigation take?

When a suspected outbreak is reported, the investigation should commence immediately and continue until there is sufficient evidence to demonstrate that the infection has been completely cleared. An outbreak of infectious disease will continue as long as there is an opportunity for transmission between susceptible and infected animals. It is the responsibility of the investigation team or investigating officer to identify and control all possible routes of transmission between infected and susceptible animals in order to control the outbreak.

You will sometimes receive reports of an outbreak after signs of disease appear to have resolved. For example, you may receive a report from a remote village stating that one month ago, 20 cattle and 5 buffalo were sick for about 10 days but because they all recovered quickly the farmers did not think it was important to report the disease. In this situation, it is important that the outbreak is still investigated and, while it may be more difficult to retrieve specific details about the outbreak after the event, the results of such an investigation can still provide useful information on possible sources of the outbreak and may help to guide preventative measures to reduce the chance of future outbreaks in the area. You may also find during your investigation that the outbreak is, in fact, still ongoing and has spread to other areas. Your

investigation will allow you to implement appropriate control measures and reduce further spread of the disease.

1.3 Who should investigate an outbreak?

As a field veterinarian or veterinary para-professional you will often be the person who first receives a report of a suspected outbreak of disease. Although you will need to inform other levels of the Veterinary Services, it is likely that you will be largely responsible for performing the outbreak investigation at the field level.

This manual outlines the approach that should be taken to investigate an outbreak. It should be stressed that it may not always be possible to follow the exact recommended steps and processes described here, due to a number of limitations, such as cost of materials and transportation, availability of personnel, and security issues preventing entry into some areas. Because of this, the material presented here should be treated as a guide. However, it should be remembered that many of the concepts and practices described in this manual can be implemented even with very limited resources. Strict biosecurity practices should always be followed by the investigating officer (see Chapter 6). You should also consider simple, often inexpensive methods of preventing transmission of disease between infected and susceptible animals. Even very simple measures can be highly effective, such as advising farmers in villages surrounding an outbreak area to avoid shared grazing and to keep cattle tethered within the household, preventing contact between infected and susceptible animals. See Chapter 2 for a description of transmission pathways (Section 2.2) and the principles of outbreak control (Section 2.6), and Appendices B.2 and C.2 for details on transmission of FMD and CSF, respectively.

1.4 The role of a disease investigator

A disease investigator is like an investigative journalist or a detective. The role of the disease investigator is to unfold the factors and circumstances leading to an outbreak. You must not only determine the animals involved in an outbreak, but also the people involved, for example visitors, animal health workers and veterinary practitioners. Investigating an outbreak of disease primarily involves gathering, recording, analysing and reporting information. You will ask many questions of different people during your investigation. The minimum questions you should ask are referred to as the 4 Ws and 1 H (see below). By asking more detailed questions, you can expect to receive more detailed answers. This is critical for getting the full picture of an outbreak.

The 4 Ws and 1 H

What? What species are affected? What age groups are affected?

When? When did the outbreak start? When did the outbreak spread to other farms?

Where? Where did the outbreak start? Where are the other farms or villages that are affected? Why? Why did the outbreak occur in this particular area?

How? How did the disease agent come onto this farm? How did the outbreak behave?

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2. Why do outbreaks occur?

This chapter will describe how and why an outbreak of disease occurs and the factors that contribute to the size of an outbreak. We will also consider how to use our knowledge of these factors to prevent or control an outbreak of disease.

2.1 Host, agent and environment

Disease is not a random event, and whether disease occurs depends upon interactions between the host, the agent and the environment in which they exist. Disease occurs when an agent capable of causing disease (for example, a virus or bacteria) meets a host that is vulnerable (susceptible) to the agent and in an environment that allows the agent and host to interact. For an outbreak to occur there must also be a chain of transmission for the agent to pass from one host to another. Whether a disease outbreak occurs will depend on factors relating to the host, the agent and the environment. The interaction between these three components is known as the disease triad, as shown in Figure 1.
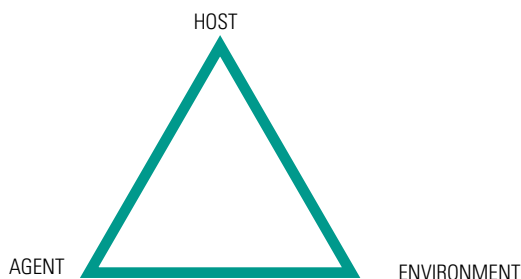
Figure 1: The disease triad of host, agent and environment

Host

A host is a living organism in which agents of disease can survive. Examples of hosts are domestic livestock such as cattle, buffalo, pigs, sheep, goats and poultry. The agent (see below) may or may not develop and reproduce in a particular host and may or may not cause disease in that host. The following host factors can determine whether disease will occur:

- Age
- Sex
- Genotype
- Behaviour
- Nutritional status
- Health status
- Immunity

Agent

An agent is the biological pathogen, such as a virus, parasite, fungus or bacterium, that causes disease in the susceptible host. An agent is primarily interested in where it can live, grow and reproduce. Some agents can survive and even multiply away from the host population (in other animals, in their products, or in the physical environment), while others must remain within the host to survive. For example, FMD virus only affects cloven-footed animals such as cattle, buffalo, sheep, goats and pigs. CSF, on the other hand, only affects pigs. The following factors determine whether an agent causes disease in a particular host:

- Infectivity
- Pathogenicity
- Immunogenicity
- Antigen stability
- Survival

Environment

The environment describes the conditions or influences that are not part of either the host or the agent, but influence their interaction. The following environmental factors can influence the occurrence of disease, provided that both the susceptible host and agent are present:

- Weather
- Housing
- Geography
- Air quality

2.2 Transmission pathways

The chain of transmission is the process by which an agent can be transmitted from a source to a susceptible host, and subsequently from one host to another. For successful transmission to occur the following factors must exist:

1. **A source of the agent**: This is often the place where the agent originates, lives, grows and multiplies. The source

(sometimes called the reservoir) of an agent can be any of the following:

– a symptomatic animal

– an asymptomatic infected animal

– an animal incubating disease

– a convalescent animal

– another animal species

– the environment

2. **A portal of exit**: The pathway by which the agent leaves the source.

3. **A mode of transmission**: The method by which the agent passes to a susceptible host. This transmission can be either direct or indirect and, for some agents, both pathways can be used.

4. **A portal of entry**: The pathway into the host, which gives the agent access to tissues where it can multiply and cause disease.

SOURCE

Cloven-hoofed animals:
– Infected and incubating disease
– Infected symptomatic animal
– Subclinically infected animal
– Infected convalescent animal
– Contaminated vehicle, clothing, etc.

PORTAL OF EXIT

FMD virus is shed in:
– Fluid from ruptured vesicles
– Expired air
– Faeces and urine
– Milk
– Semen

Direct spread:
Direct contact between infected and susceptible animals

Direct

MODE OF TRANSMISSION

Indirect

Indirect spread:
– Vehicles
– Clothing, people, vets
– Other animals
– Feed, manure, equipment

PORTAL OF ENTRY

Portals of entry include:
– Inspiration
– Ingestion
– Skin or mucous membrane abrasions

Figure 2: Transmission pathway example — foot-and-mouth disease.

2.3 When does an outbreak occur?

As described above, for a disease outbreak to occur there must be an agent, a susceptible host and a means by which the agent can be transmitted from an infected host to subsequent susceptible hosts. When an outbreak occurs at a particular time, it is because of a change in the natural 'balance' between host, agent and environment. Any one of the following changes could result in an outbreak:

– **An increase in exposure to the agent**. For example, the amount of agent can be increased to a level capable of causing infection if environmental conditions favour survival of the agent.

– **An increase in the infectivity/virulence of the agent**. For example, changes to the agent's genotype can occur in which the virulence of the agent can be increased, overcoming resistance of the host and resulting in disease.

– **xposing the agent to naïve hosts**. For example, a cow is brought from an infected area into a village where there has been no disease. When she arrives at the village, she might not be showing any signs of illness, but could be incubating disease (infected but not yet showing clinical signs). This cow would then introduce infection into the village and could infect other animals.

– **An enhancement of transmission**. For example, if there is greater opportunity for the agent to be moved between infected and susceptible animals.

– **Increased host exposure**. For example, the host and agent may have both been present, but an outbreak occurs when the host has greater exposure to the agent.

– **Change in susceptibility of host to the agent**. An outbreak can occur when the susceptibility of the host to an agent changes. For example, animals previously vaccinated or exposed to FMD virus will develop immunity; however, in time, this immunity will wane and the host will again become susceptible. Young or naïve animals may also be introduced into a population. An outbreak will occur when there are sufficient numbers of susceptible animals.

– **Introduction through new portals of entry.**

2.4 The host population

We have already established that for an outbreak to occur there must be adequate numbers of susceptible hosts as well as an agent, a suitable environment and means by which the host and the agent can come together. Here, we will consider in more detail the role of the host population. The proportion of susceptible animals in a population is a major determinant for whether an outbreak will occur and the evolution of that outbreak. In an outbreak of infectious disease, individuals in the population can be classified into one of the following groups:

– Susceptible

– Incubating

– Incubating and infective

– Diseased and infective

– Sub-clinical and infective

– Convalescent and infective

– Convalescent

– Dead

– Immune
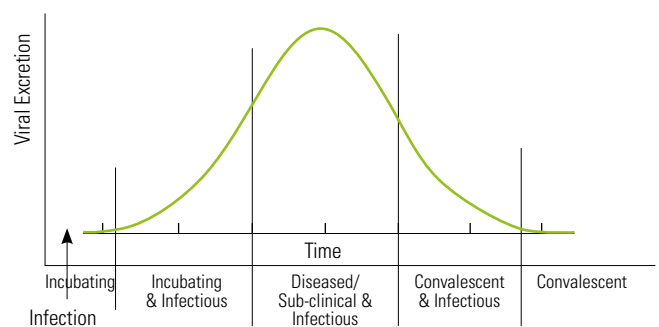
Figure 3: Line plot showing the relative amount of viral excretion from an infected host, as a function of time since infection.

An individual's vulnerability to infection, ability to infect others, and risk of being infected will depend on which of the following categories it belongs to:

1. **Susceptible animals**. Susceptible animals are those individuals in the host population that are vulnerable to infection by an agent. The proportion of susceptible animals in a host population will influence whether an outbreak of disease will occur and the extent of the outbreak.

2. **Incubating animals**. The incubation period of a particular disease is the period of time between infection of an animal and development of clinical signs. An incubating animal is infected with the agent but is not

showing any signs of infection. With some infectious diseases (such as FMD) an infected animal starts to shed the agent before it starts to show clinical signs.

3. **Diseased animals**. Diseased animals are those that are showing clinical signs. With most infectious conditions (not all) those that are showing clinical signs are also shedding the infectious agent and therefore pose an infectious risk to other susceptible animals.

4. **Sub-clinical animals**. Sub-clinically infected animals are those that are infected with an agent, are capable of transmitting the agent to susceptible hosts, but do not show clinical signs of disease themselves. These animals can be highly infectious. In a similar way to incubating animals, sub-clinical animals represent an important risk, because they are a source of infectious agent but are not easily detectable. Sub-clinical infections are known to occur with FMD, and there have been instances in which outbreaks have resulted from the introduction of a sub-clinically infected animal into a susceptible population.

5. **Convalescent animals**. Convalescent animals are those that have recovered from disease and no longer display clinical signs. Some convalescent animals will continue to shed infectious agent in the early stages of recovery and, therefore, represent a risk to susceptible animals. Most will only shed virus for a short time, or not at all, after clinical signs have resolved. For FMD, some animals become carriers, with virus able to be isolated from them for at least 28 days after recovery. Although this can happen, these animals are not thought to be important sources of transmission of virus to susceptible hosts.

6. **Immune animals**. Immunity can occur in a number of ways: following infection, following vaccination or, in the very young, due to the presence of circulating maternal antibodies. The period of immunity varies depending on the agent involved and depending on whether it is due to natural infection or to vaccination. For FMD, immunity following natural infection is variable and can last up to two years. Following vaccination, it is generally understood that protective levels of immunity will be maintained for anywhere between 6 and 12 months. Therefore, in the case of FMD, the immune period is transient and animals (and indeed a population) will again become susceptible if they are not exposed to the agent (through either infection or vaccination) on a regular basis. The introduction of new animals to a population, either through birth or replacements from unaffected areas, increases the proportion of susceptible animals in a population. A high proportion of susceptible animals will increase the likelihood of an outbreak, if an infective agent is introduced.

2.5 Epidemic curves and the reproductive ratio

Epidemic curves

An epidemic curve is a frequency histogram showing the count of new cases of disease (on the vertical axis of the plot) as a function of time (on the horizontal axis). Epidemic curves are useful because they show the progression of an epidemic (or outbreak) over time. The shape of the curve can provide useful indications about the nature of disease transmission and the likely stage of an outbreak. The following factors will influence the shape of an epidemic curve:

– The incubation period of the disease

– The infectivity of the agent

– The proportion of susceptible animals in the population

– Animal density (which influences transmission of infective agent from one animal to another).

For example, a highly infectious agent with a short incubation period infecting a population with a high density of susceptible animals produces an epidemic curve with a steep initial slope over a relatively short timescale, representing a rapid spread of infection among the population.

Common-source epidemics

In a common-source epidemic (or outbreak), a group of animals are exposed to a common source of infective agent. If the group is exposed over a relatively short period, disease cases will emerge over one incubation period. This is called a common point source epidemic. With a common point source epidemic, the epidemic curve rises rapidly and contains a definite peak at the top, followed by a gradual decline. Figure 4 illustrates an example of a common source epidemic.

> Delivery of a consignment of feed contaminated with toxin from Clostridium botulinum to a feedlot would produce an epidemic curve consistent with a common point source epidemic (assuming the contaminated material was fed out to stock over a relatively short period of time).

Exposure can also occur over a longer period of time, either intermittently or continuously. This creates what is known as a continuous common-source epidemic. This epidemic curve rises rapidly (associated with the introduction of the agent). After the initial rise, the number of cases plateaus rather than tapering off.

Propagated epidemics

A propagated epidemic occurs when a case of disease serves as a source of infection for subsequent cases and those subsequent cases, in turn, serve as sources for later cases. An epidemic curve of an uncontrolled outbreak of FMD would produce an epidemic curve typical of a propagated epidemic. In theory, the epidemic curve of a propagated epidemic has a successive series of peaks reflecting increasing numbers of cases in each generation. The epidemic usually wanes after a few generations, either because the number of susceptible animals falls below a critical level, or because intervention measures become effective. Figure 4 illustrates an example of a propagated epidemic.

Uncontrolled spread of FMD in a population of livestock farms would produce an epidemic curve consistent with a propagated epidemic.
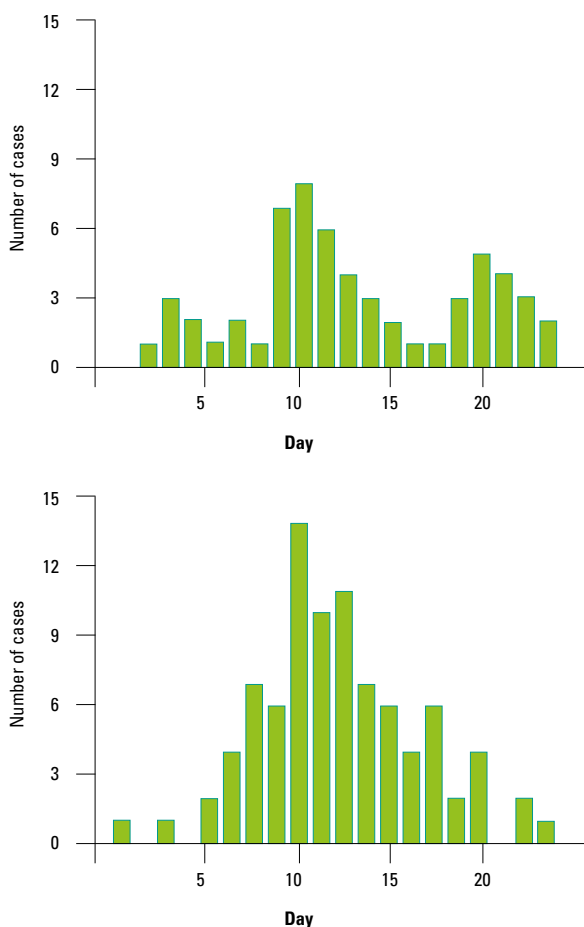
Figure 4: Epidemic curves. The plot on the left is typical of a propagated epidemic. The curve on the right is typical of a common-source epidemic.

Estimating the time of exposure

In a common point source epidemic of a known disease with a known incubation period, you can use the epidemic curve to identify the likely period of exposure. To identify the likely period of exposure from an epidemic curve:

– Determine the average, minimum and maximum incubation period for the disease of interest. For FMD these intervals are 4 days, 2 days and 14 days (respectively). For CSF these intervals are 8 days, 5 days and 10 days (respectively).

– Identify the peak of the outbreak or the date on which the median (i.e. middle) case was observed and count back on the horizontal axis one incubation period. Make a note of this date.

– Start at the earliest case of the epidemic and count back the minimum incubation period. Make a note of this date.

Ideally, the two dates will be similar, and will identify the time range over which exposure occurred. However, this technique is not precise, and you usually should widen your period of investigation by

10% to 20% on either side of these dates. You should then investigate possible exposures during this wider time frame in an attempt to identify the possible source.

Sometimes, the endpoint of an epidemic is difficult to pinpoint. A way of getting around this problem is to do a natural log transformation of the horizontal axis (time), which conveniently normalises the (typically) skewed epidemic curve. With the epidemic curve normalised, you can now use start time, end time, and the date of the median case to calculate the incubation period. Although transforming the data to the logarithmic scale gives it convenient mathematical properties, it is important to transform the values back to their original scale before making interpretations.

Interpreting the epidemic curve

The first step in interpreting an epidemic curve is to consider its overall shape. The shape of the epidemic curve is determined by the epidemic pattern (common source versus propagated), the period of time over which susceptible individuals are exposed, and the minimum, average, and maximum incubation periods for the disease. An epidemic curve which has a steep upslope and a more gradual downslope (a log-normal curve) indicates a common point source epidemic in which individuals are exposed to the same source over a relatively brief period. In fact, any

sudden rise in the number of cases suggests sudden exposure to a common source. In a common point source epidemic, all cases occur within one incubation period. If the duration of exposure was prolonged (a continuous common-source epidemic) the epidemic curve will have a plateau instead of a peak. Intermittent common-source epidemics produce irregularly jagged epidemic curves which reflect the intermittency and duration of exposure and the number of individuals exposed. Propagated epidemics, in theory, show a series of progressively taller peaks one incubation period apart, but, in reality, few produce this classic pattern.

Cases that stand apart may be just as informative as the overall pattern. An early case may represent a background or unrelated case, a source of the epidemic, or an individual who was exposed earlier than most of the cases. Similarly, late cases may represent unrelated cases, long-incubation-period cases, secondary cases, or individuals exposed later than most of the cases. Sometimes, these outliers represent miscoded or erroneous data. All outliers are worth examining carefully, because if they are part of the outbreak, their unusual exposures may point directly to the source.

You can also use the principle of the epidemic curve to determine whether emergency control measures have been successful. If measures such as movement controls (preventing shared grazing, tethering animals within the household area) or emergency vaccination have been successful, there should be a decline in the number of new cases following the date on which the control measure was established. However, this decline will not be immediate because those animals that were infected before control measures were implemented will still develop infection even after the controls have been implemented. Therefore, you would still expect to see a decline in new clinical cases approximately 1 – 4 days (the most common incubation period for FMD) after implementation of effective control measures.

The basic reproductive number

The basic reproductive number (R_0, 'R nought') is the average number of secondary cases an infectious individual will cause in a completely susceptible population. R_0 provides a measure of the intrinsic potential of an infectious agent to spread. R_0 depends on a number of factors:

– The number of contacts made

– The probability of infection given successful contact

– The duration of infectiousness

– The proportion of contacts that are susceptible

If R_0 for an infectious disease is less than one, it is unlikely that an epidemic (or outbreak) will occur if the agent is introduced into a susceptible population. If, on the other hand, R_0 is greater than one, introduction of the infective agent into a susceptible population is likely to produce an epidemic. The larger the R_0 value, the more rapid the spread of disease.

As an infectious agent invades a population, the number of susceptible animals progressively declines as a result of either recovery or death. Eventually, insufficient susceptible animals are present to maintain the chain of transmission. At this stage, on average, each infectious animal infects less than 1 susceptible animal and the epidemic dies out. This is explained in Figure 5.
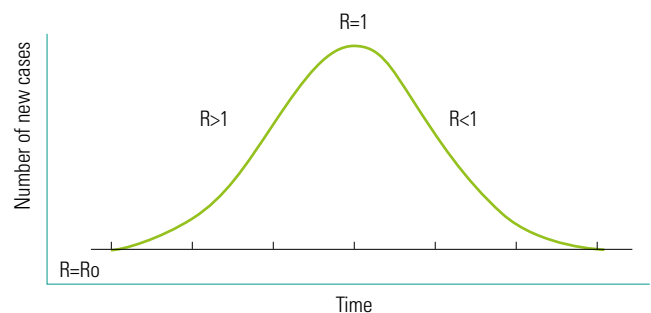
Figure 5: Reproductive number according to the stage of an epidemic.

The estimated dissemination ratio

A metric called the estimated dissemination ratio (EDR) can be used to provide an estimate of R_0 during an infectious disease outbreak.

The n-day EDR at day i of an outbreak equals the total number of incident cases between day i and day $[i - (n - 1)]$ divided by the total number of incident cases between day $(i - n)$ and day $(i - 2n)$. EDR values are often calculated for each day of an epidemic and presented as a time series. If the EDR is consistently less than one, the epidemic is said to be 'under control'.

Figure 6 is an epidemic curve showing the temporal evolution of an infectious disease outbreak in poultry. Superimposed on this (as a line) is the 4-day EDR plot. If we are on day 30 of the epidemic, the 4-day EDR value for day 30 equals the number of FMD cases identified between day 27 and day 30 divided by the number of cases identified between day 23 and day 26. So, if the number of cases in the 27 to 30 day window is greater than the number of cases in the 23 to 26 day window, our EDR value (an estimate of R_0) is greater than one.
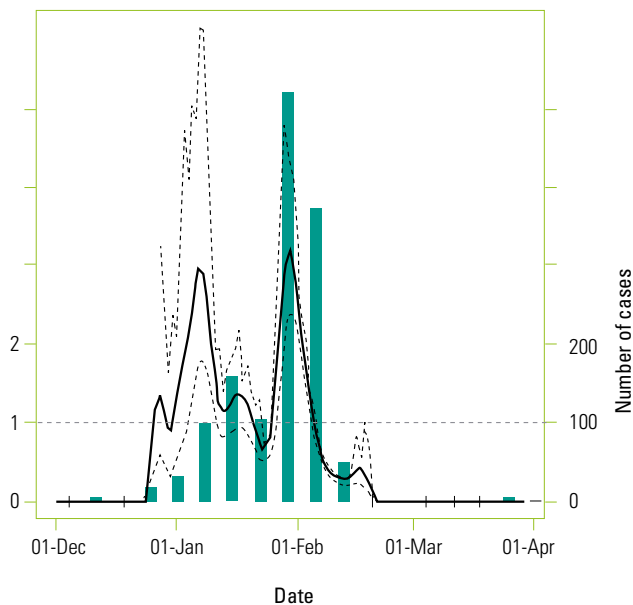
Figure 6: Data from an infectious disease outbreak in poultry. Histogram of counts of cases as a function of calendar date. Line plot showing estimated dissemination ratio (and its 95% confidence interval) as a function of calendar date.

EDR plots are particularly useful for infectious disease outbreak investigations because they provide warning that an outbreak has not yet reached its peak. In Figure 6, the relatively high EDR values (greater than 1) in early March 2001 indicated that the epidemic had not yet reached its peak. By the time the epidemic had reached its peak in early April 2001, the EDR plot had started to drop below one, indicating that the epidemic had largely been brought under control. From May 2001 the 4-day EDR plot fluctuated around one, indicative of ongoing disease transmission (evidenced by the long 'tail' to the epidemic).

2.6 Principles of outbreak control

In this chapter we have discussed how and why an outbreak develops and what factors influence disease incursion and spread. We will now consider how to use this knowledge to either prevent or control an infectious disease outbreak. The principles of disease control involve the following:

1. **Reducing contact between infectious and susceptible animals**. Intervention measures to reduce contact between infectious and susceptible animals include separation or quarantine of infected animals, and movement controls. It is also important to avoid mixing of livestock when an outbreak is occurring, for example: sharing common grazing areas with cattle from a village affected by FMD should be avoided.

2. **Reducing the number of susceptible animals**. As described above, for an outbreak to occur, there must be sufficient susceptible animals in the population to allow for continued transmission of the agent from infected to susceptible animals. By reducing the number of susceptible animals, the extent of the outbreak will be reduced. If the number of susceptible animals can be reduced to below the threshold level, an outbreak will not occur. Vaccination is an effective way to reduce the number of susceptible animals in a population.

3. **Decreasing the amount of infectious agent**. Decreasing the amount of infectious agent reduces the level of agent the host is exposed to or prevents exposure completely. This type of measure is usually aimed at a point in the transmission pathway when the agent is at its most vulnerable. For example, during the time that the agent is exposed to the external environment, it is highly vulnerable to human intervention. An example of this type of intervention would be application of disinfectants to contaminated areas or contaminated equipment.

3. A step-by-step guide to outbreak management

This section and subsequent sections of this document form the basis of the step-by-step approach to outbreak investigation and management. Figure 7 provides an outline of the steps that comprise the outbreak investigation and management process. The previous sections of this document provided background information about factors influencing how disease spreads in animal populations. The subsequent sections provide specific details about various aspects of infectious disease outbreak management, based on the steps shown in Figure 7.
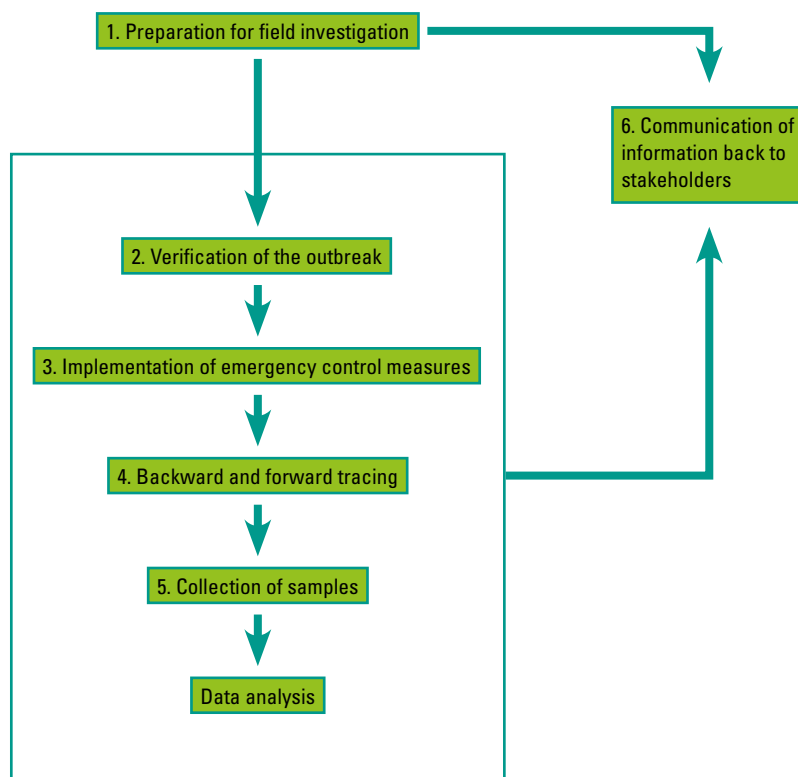
Figure 7: Diagrammatic representation of an approach to outbreak investigation and management.

4. Gathering information and preparing for the investigation

This step covers the period from when you first receive a report of a suspected outbreak to when you begin your field investigation. It will consider how you are likely to hear of a disease outbreak; how you should record this initial report; and how and what you should prepare before embarking on a field outbreak investigation.

4.1 Receiving a report and collecting initial information

A report of a suspected outbreak can be made by a number of different people, for example:

– Farmers

– Village animal health and veterinary workers

– Local authorities

– Members of the public

– Private veterinarians

– Livestock traders

– Medical doctors (in the case of zoonotic diseases)

– Others

Reporting from any of these groups should be encouraged and any report received should be taken seriously and followed up. Issues of outbreak detection sensitivity and specificity are important here. If your outbreak detection criteria are too **sensitive** (that is, the criteria of what constitutes an outbreak are too liberal) it is likely that a considerable number of the outbreaks you investigate will

be false positives. If your outbreak detection criteria are too **specific** (that is, the criteria of what constitutes an outbreak are too strict) it is likely that all of the outbreaks that you investigate will be true positives (that is, you will not waste any time investigating false positives), but in doing so it is likely that you will miss a potentially important number of true positives.

> As a rule of thumb, the same or similar report of an unusual pattern of disease events from one or more independent sources provides strong evidence that the reported pattern of disease events is a true-positive (as opposed to being a false-positive) disease report.

Failure to act on a report may discourage future reporting of disease, resulting in an outbreak going unnoticed and uncontrolled.

At the time that a disease report is first received, some basic information should be retrieved so that there is a record of the report for future reference. At this time, you can collect some of the main details of the event to help guide your initial response. Such information can be collected in the office log book, diary or other recording system used within the office. The entry in the book might look similar to Figure 8.

A small amount of information (such as in the example above) can provide valuable clues about the nature of the disease, location, and the extent and time frame of the outbreak. A lot more information will be required later in the investigation when you go to the field, but a brief description such as this is a valuable guide for your initial response.

DATE	NAME OF REPORTING PERSON	ADDRESS/location OF OUTBREAK	DESCRIPTION OF COMPLAINT
12th December 2008	Min Kyaw Oo	Shetwa village kalwin village Track	3 Cattle lame last 2 days. Not eating, heard neighbour had 1 sick cow

Figure 8: An example of a log-book entry showing details of a report received on a suspected outbreak. It includes the farmer's name and address and a brief history of the event.

4.2 Preparing for a field outbreak investigation

Before starting the field component of the outbreak investigation, you should ensure that you are well prepared for field activities. This involves making sure you have all the equipment and information you require to conduct the initial outbreak investigation.

Preparation of equipment

It is important that you take with you the essential equipment needed to conduct an outbreak investigation in the field. This may vary a great deal in each situation. However, there are certain items which should always be included, such as disinfection equipment (to ensure that you can at least disinfect your own vehicle and equipment after leaving an infected area to avoid transmitting disease to other areas), recording equipment, e.g. paper and pens and/or a specific form for taking down information, and animal restraint equipment to ensure that you can examine animals safely and effectively.

An equipment checklist is provided in Appendix D as a guide to the equipment needed for a field investigation. The checklist can also be used at times when there is no

outbreak. As part of emergency response preparedness, a checklist is important to ensure that you have everything on hand at the (often unexpected) time you need it.

Preparation of information

Before embarking on a field visit, it is important that you equip yourself with certain information that will help you during your investigation. The information provided to you at the time the outbreak is reported (for example, Figure 8), should already provide details about the species of animals involved, the time of the outbreak, the clinical signs, and the location of the outbreak. Taking the example from Figure 8, we will consider what can be learned from even a small amount of information provided in the initial report. Figure 9 provides annotations to the log-book entries shown in Figure 8 to show how even very basic outbreak information can be used to guide further outbreak management activities.

Information you should have before investigating an outbreak

1. **Location**. You should know the location of the outbreak from the initial report. Before going to investigate the outbreak you should find out more information about that place. Have there been any previous outbreaks in the area? Are any outbreaks ongoing? Which livestock

Figure 9: An example of a log-book entry of an initial report of an outbreak and how a simple record can provide valuable information to assist in preparations for an outbreak investigation.

species and husbandry methods are used? Which main roads/transport routes are used? Where are animals and animal products obtained from? Where do they go once they are sold? Are there any livestock markets and slaughterhouses that might be relevant to the outbreak? By knowing the location of the outbreak, you can also prepare maps of the area, which are often very useful at the time of the investigation.

When you travel to the outbreak area, make sure that you have with you the facilities to record details of outbreak locations. Facilities, in this sense, would include a GPS-enabled smartphone and detailed maps of the area (which would allow you to record the exact location of affected villages or households).

2. **Disease information.** If you have an idea of possible disease processes or diagnoses based on the information that has been provided, it is important that you have a good working knowledge of the aetiology, epidemiology, pathogenesis, clinical signs, diagnosis, treatment and control of these diseases. Some of these details for FMD and CSF are provided in the appendices. This information will help you to implement suitable emergency control measures and provide advice to farmers and other livestock owners.

Once you have prepared the equipment that you require and you have all the available information on the location and the suspected diseases, you can then commence the field component of the outbreak investigation.

5. Verifying that you actually have a problem

Verifying that you actually have an outbreak is the first step that occurs in the field. Once you have verified that an outbreak exists, you should then proceed to:

1. Identify the possible causes of the outbreak.

2. Make an initial assessment of the extent of the outbreak.

3. Gather information to guide further investigation and control measures.

Information gathering once you arrive at the outbreak farm or village will allow you to develop a list of differential diagnoses, to understand the timing and extent of the outbreak and to identify the index case. Identification of the index case and all cases prevalent at the time of initial investigation is important because it allows you to identify the likely source of infection and where disease has spread. Often, you will have a suspicion of the disease causing the outbreak at this stage and it will be necessary to implement control measures based on your assessment of the situation. Rapid implementation of control measures, even before a definitive diagnosis has been reached, will help to limit the spread and the size of the outbreak. This section is divided into two parts, based on the information collected during this phase of the investigation.

5.1 Gathering information from stakeholders

It is important during this stage of the investigation that you collect core information from individuals closely involved with the affected animals, including farmers, livestock owners and livestock traders. An outbreak investigation form (Appendix A) can and should be used to guide the questioning process during this phase of the investigation. However, you should understand the key questions and the objectives of this step, so that you can conduct interviews and information-gathering exercises even when you are called unexpectedly to an outbreak and do not have the form with you. The information that needs to be collected can be divided into three main categories:

1. Animal: details of the animals involved in the outbreak (species, breed, age).

2. Place: details of the geographical location of disease-positive and disease-negative households, farms or villages.

3. Time: details of the time of onset of disease in affected households, farms or villages.

Further details and a more complete description of important questions to ask are provided below.

Animal

Animal factors are consistent with the description of the 'host' described in Chapter 2. Animal factors include details of the species, age and sex of disease-positive ('affected') and disease-negative ('non-affected') animals in the population at risk. Enumerating the number of affected animals and comparing that number with the size of the population at risk (that is, calculating an attack rate) and then comparing attack rates for different classes of stock (e.g. different species, different ages, different breeds) provides valuable information.

There is some basic information that must always be collected during an outbreak investigation:

1. Counts of the animal population at risk of disease. Ideally this will be categorised into counts of individuals by species and age.

2. Counts of the number of affected animals by species and age (*morbidity*).

3. Counts of the number of dead animals by species and age (*mortality*).

This basic information will often be recorded using an outbreak investigation form. If the form is not available, you can easily record the information in your notebook in the format shown in Table 1.

Table 1: An example of recording animal information during an outbreak using a simple table, where an outbreak investigation form is not available.

Location	Total	Sick	Dead
Farm 1 - cattle	3	3	0
Farm 1 - buffalo	1	0	0
Farm 1 - pigs	2	0	0
Farm 1 - goats	0	0	0

Attack rates

When describing the frequency of disease in animal populations we use the concept of attack rates. The attack

rate is the number of cases of a given disease event divided by the number of animals at risk at the beginning of an outbreak (in epidemiology, 'attack rate' and incidence risk [cumulative incidence] are synonymous). The attack rate provides a quantitative measure of the proportion of the at-risk population that are disease-positive.

$$\text{Attack rate} = \frac{\text{Number of cases of disease}}{\text{Number of individuals at risk}} \quad (1)$$

In addition to calculating a point estimate of attack rate, we also need to be aware of the certainty that we have about that point estimate. A measure of certainty comes from calculating a confidence interval around the point estimate. The point estimate of attack rate makes a statement of our best estimate of the true population value of the frequency of disease in a given population at a given point in time. A confidence interval around that point estimate provides the likely range of values we expect that population value to take. For example, a 95% confidence interval for a given attack rate provides the minimum and maximum bound of the attack rate values that encompass 95% of the true population values.

The confidence interval for a proportion (i.e. an attack rate) is calculated as follows:

$$\hat{p} \pm (z \times SE_p) \text{ where } SE_p = \overline{\sqrt{\frac{\hat{p}(1-\hat{p})}{n}}} \quad (2)$$

Where $\hat{p}$ is the observed proportion, n the sample size and z the appropriate critical value from the z distribution (use $z = 1.96$ if you are calculating a 95% confidence interval).

Consider an outbreak of FMD in a village where there are 100 cattle and 30 buffalo. Of this group, 20 head of cattle and 3 head of buffalo are identified as FMD-positive. The attack rate would be:

Attack rate = 23 ÷ 130

Attack rate = 0.18

Attack rate = 18 FMD cases per 100 animals at risk.
The 95% confidence interval for the attack rate is:

$SE_p = [(0.2 \times (1 - 0.2)) / 130]^{0.5}$

$SE_p = 0.035$

Lower bound of 95% CI = 0.2 − (1.96 × 0.035)

Lower bound of 95% CI = 0.13

Upper bound of 95% CI = 0.2 + (1.96 × 0.035)

Upper bound of 95% CI = 0.27

The FMD attack rate in this village is 18 (95% CI 13 to 27) cases per 100 individuals at risk.

It is often useful to compare attack rates for different classes of stock:

Attack rate for buffalo = 3 ÷ 30

Attack rate for buffalo = 0.10

Attack rate for buffalo = 10 FMD cases per 100 animals at risk.

Attack rate for cattle = 20 ÷ 100

Attack rate for cattle = 0.20

Attack rate for cattle = 20 FMD cases per 100 animals at risk.

The FMD attack rate for buffalo is 10 (95% CI 4 to 26) cases per 100 individuals at risk. The FMD attack rate for cattle is 20 (95% CI 13 to 29) cases per 100 individuals at risk. This suggests that cattle are twice as likely to be identified as FMD-positive, compared with buffalo. This example shows how attack rates can be useful for comparing the frequency of disease in different groups of animals. A similar process can be followed for sex, age or other variables.

When using measures of disease frequency, you can see from the example above that we always include details on the number of animals affected and the total number of individuals at risk. If we do not account for the size of the population of individuals at risk we cannot make a valid comparison across groups. For example, if we go to a village and find that 30 young cattle (less than 24 months of age) and 30 older cattle (greater than 24 months of age) have FMD, we cannot make a valid comparison of the frequency of disease across the two age groups because we have no idea of the total number of animals present in each age group.

If it is established that there are 30 young cattle and 150 older cattle in the village, attack rates for the two groups would be calculated as follows:

Attack rate for young cattle = 30 ÷ 30

Attack rate for young cattle = 1.00

Attack rate for young cattle = 100 (95% CI 89 to 100) FMD cases per 100 animals at risk.

Attack rate for older cattle = 30 ÷ 150

Attack rate for older cattle = 0.20

Attack rate for older cattle = 20 (95% CI 14 to 27) FMD cases per 100 animals at risk.

Our analyses indicate that younger cattle are five times more likely to be affected by FMD than older cattle. This might be due to increased immunity in the older animals due to exposure to virus from a previous outbreak or due to vaccination. Alternatively, it could be due to other factors

such as management differences or behaviour differences between younger and older cattle.

Some diseases have very obvious clinical signs and, if they are present, arriving at a presumptive diagnosis is quite straightforward. For example, if vesicles are observed on the mouth and/or on the feet of cattle, buffalo and/or pigs then you would have a very strong suspicion of FMD or other vesicular diseases.

If less-specific signs are observed, such as fever, lethargy and inappetance, it may not be possible to come to a provisional diagnosis based on clinical signs alone. However, if a number of animals are affected with these signs, this should be enough to prompt implementation of control measures in order to prevent further spread of disease. Further investigation can continue once control measures have been put in place. Veterinarians and veterinary para-professionals should be familiar with the clinical signs of the major (important) infectious diseases of livestock in their area. Appendices B and C of these notes provide summary information on the aetiology, epidemiology, clinical signs, and diagnosis of FMD and CSF, respectively.

The following are provided as examples of the type of questions you might ask in order to understand better the animal factors in an outbreak situation. This list is not exhaustive and you are encouraged to add more questions depending on the situation and your experience.

– What species are affected?

– What are the clinical signs observed?

– How many animals are affected in the farm/village?

– How many susceptible animals are there in the household, farm or village?

– What is the age of affected animals?

– What is the gender and stock class of affected animals: male, female or both?

– What is the management system in the village: free grazing, tethered in household, commercial farm, etc?

– In which animals (species, age, sex) were clinical signs first observed?

Time

There are three basic time spans used to describe the temporal pattern of disease:

1. **Epidemic period.** A period of variable length depending on the duration of a particular outbreak.

2. **Seasonal trends**. A 12-month period used often to describe the cyclical pattern of disease that varies according to the time of the year.

3. **Long-term trends.** An indefinitely long period (often years) used to identify patterns of disease over extended periods of time.

The following list provides examples of some questions that might be asked relating to the time factor of an outbreak. Again, this list is not exhaustive.

– When did the outbreak start?

– When were clinical signs first seen?

– When was the last outbreak of this disease, or similar clinical signs seen in this household, farm or village?

– How often have you experienced an outbreak like this one?

– On which household, farm or village were clinical signs first noticed?

– In which animal were clinical signs first noticed?

Place

Describing an outbreak in terms of place can yield important information about the cause of an outbreak. For cattle in a feedlot situation, we might look at the pattern of the outbreak among different pens or buildings of the feedlot. More often, however, we would be considering patterns on a larger scale, such as different households or farms within a village, or villages within a village tract.

It is often useful to consider place and time together. A useful way of documenting place and time is to draw a plan of the layout of the village (or farm, or village tract) and record on that map the dates on which clinical signs were observed. Such a diagram can provide important information on the pattern of the outbreak. It is often helpful to sketch this type of diagram during your visit to the affected area. Figure 10 shows a diagram of a village affected by FMD.

A diagram similar to that shown in Figure 10 can be constructed during an outbreak investigation in order to guide further questioning about the source and spread of the outbreak. In order to construct such a diagram, the following questions could be asked:

– What area is affected by the outbreak — a feedlot, village, village tract?

– In which farm were clinical signs first observed?

– At each farm (or pen, or village) ask what date were clinical signs observed in that place.

– Is the affected farm or village near to a livestock market or slaughterhouse?

– Have outbreaks of disease like this been experienced in this place before?

5.2 Putting it all together

We have now covered the concepts of collecting information about animal, time and place in order to understand outbreak patterns. We have outlined some questions that can be asked during the investigation, noting that the questions provided can be expanded to gather more information. It is important to use an outbreak investigation form to guide the interview process and information-gathering procedures, but the investigator must also be equipped with a sound working knowledge of the important infectious diseases of livestock in his/her area so that investigations can be implemented quickly if and when they are necessary.

Now that we have established the type of information to collect, we should consider methods of collecting information. On arrival at the village, you are likely to meet with the local authority, the village head and the farmer who reported the outbreak, along with other farmers and stakeholders. How you proceed to collect information will depend on the local situation and customs in a given area. It may not be suitable to use an outbreak investigation form while retrieving information from these people, as they might become worried about the information being reported in an official document. Sometimes, you will need to collect the information through discussion, make some rough notes and then complete the form later. There are different ways of gathering information in the village setting. Further details on how to organise and conduct these meetings is provided in Chapter 10. Here, we look at just two examples of how information might be collected from stakeholders in the field.
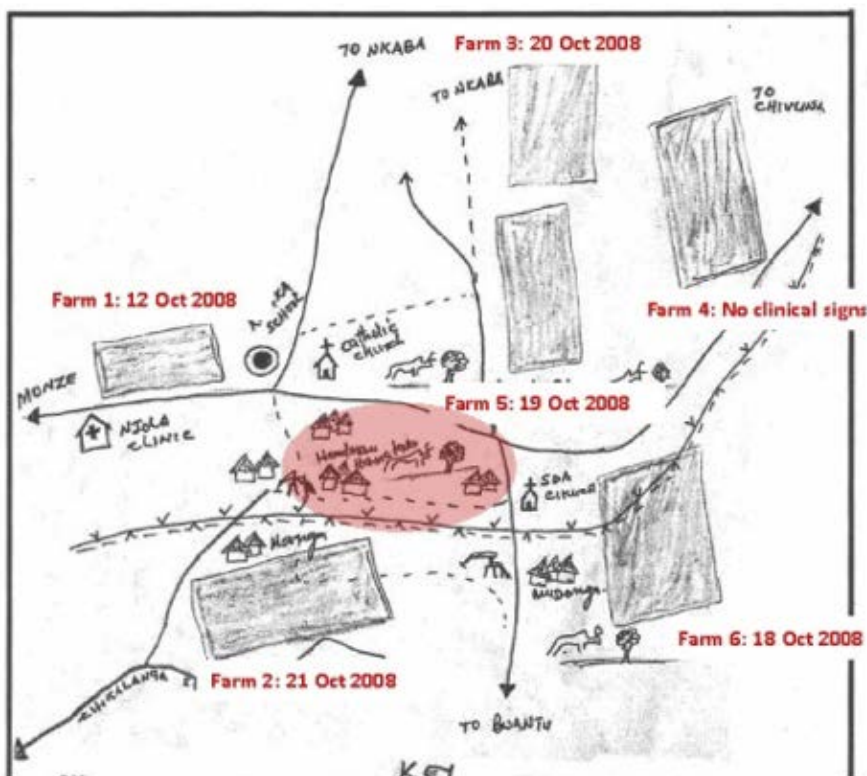
Figure 10: A sketch of the outbreak area (constructed with the assistance of local livestock owners) is a useful way to describe and understand the spatial distribution and timing of disease events. In this example, we can see that Farm 1 was the first place where clinical signs were seen. Therefore, Farm 1 is the index case for the outbreak in this village. By knowing when disease occurred on each farm, we can understand better how the disease might have been transmitted between farms. A diagram such as the one above will help to prompt further questioning. For example, does Farm 1 use the common grazing area (indicated by the red-shaded area) and, if so, which other farms use this grazing area?

A group meeting such as a Dutaik[1] meeting (see Figure 11) to discuss the outbreak with a number of farmers and also other people from the village can often be useful for collecting outbreak information. At such a meeting, farmers might share their experience of past outbreaks similar to the current one. A village meeting provides a good opportunity for you to discuss with stakeholders how you will proceed with the investigation and the control measures that will be implemented. Ensuring that all stakeholders are well informed is one way of ensuring that control measures will be conscientiously followed.

Figure 11: Small group discussions or individual discussions can be useful for gathering information.

Sometimes, you may interview a number of farmers individually in order to get detailed information on the outbreak occurring in their animals, or you may organise small group discussions between a number of farmers, perhaps where there are a number of neighbouring farms affected.

While you are gathering information on animal, time and place during this step, it is important that the information you collect is accurately and consistently recorded (see Figure 12). Filling in an outbreak investigation form for each household, farm or village affected provides a convenient means for doing this. If you do not have the outbreak investigation form with you at the time of the outbreak, some simple and effective methods can be used for recording key information. This information might be collected into a note book or into your office log book. The details recorded for each household, farm or village include:

1. The name of the person responsible for the care of the affected stock.

2. The location of the household, farm or village.

3. The species of animal affected.

4. The total number of susceptible animals.

5. The number of affected animals and number of dead animals.

6. The time when clinical signs were first observed.

7. A description of the clinical signs.

Clinical examination of affected animals is an important part of the outbreak investigation process. The veterinarian or the veterinary para-professional should wear appropriate protective clothing and they should be able to safely restrain affected animals, conduct a thorough clinical examination and then make an assessment of the similarity of the observed clinical signs with those of the important infectious diseases of livestock such, as FMD and CSF (Appendix B and C).

In order to ensure that biosecurity remains paramount, you should set up two types of outbreak investigation teams: one team that will investigate known infected areas, and another team that will go to areas still considered to be free of disease. The purpose of the second group is to conduct active outbreak investigation to search for unreported cases of disease. Use of a 'clean' investigation team means that there will be no way that veterinary staff can be responsible for introducing infection into areas that are free of disease.

At the same time that you examine animals in an affected area, samples should be taken to aid diagnosis and to provide further epidemiological information on the outbreak. This may include blood and/or serum samples but, in the case of CSF suspicion, a post-mortem examination may be required. If a post-mortem is performed, appropriate tissue samples should be taken for virus isolation.

Figure 12: Small group discussions or individual discussions can be useful for gathering information.

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1 A Myanmar term for a community gathering where people sit in a circle with their knees touching each other's

6. Implementing emergency control measures

6.1 Principles of control measures and biosecurity

One of the major objectives of investigating an outbreak of disease is to control the spread of disease and then to eradicate disease as quickly as possible. A rapid outbreak response and the swift implementation of appropriate and effective control measures are vital for successfully controlling disease. This may involve introducing very simple control measures, such as preventing animals in outbreak areas and surrounding areas from using shared grazing areas. It could also involve keeping susceptible, as well as infected, animals confined to their household to minimise the risk of contact between infected and susceptible animals.

Biosecurity is vital in containing an outbreak. Biosecurity measures are the measures taken to reduce the chance of transmitting infection from an infected area (or animal) to an unaffected area (or animal). This includes the measures applied on the farm itself (both affected farms and unaffected but at-risk farms) and the measures applied when leaving/entering the farm. Veterinarians, veterinary para-professionals and any other personnel involved in the outbreak investigation will often disinfect their footwear, change their clothes and bathe before leaving an infected area, and will disinfect their vehicles and equipment both when leaving an infected area and entering an unaffected area.

The central concept and objective of control measures and biosecurity is to prevent transmission of infection from an infected animal to a susceptible animal, and therefore break the transmission pathway (see Chapter 2 on why an outbreak occurs). There are a number of ways in which the transmission pathway can be interrupted, but in order to do this effectively, it is necessary to understand certain characteristics of the disease agent involved in the outbreak. The following information should be known:

1. **Transmission dynamics/transmission pathway**. It is important to understand how a disease agent is transmitted from an infected animal to a susceptible animal. Transmission can be direct, indirect or vertical (from parent to offspring).

2. **Survival of the infective agent in the environment.** It is important to know how the causative agent survives in the environment. The environmental stage of a disease agent's life cycle is often its most vulnerable, and is therefore easily targeted by intervention measures. For example, FMD virus is sensitive to pH and can be easily killed using appropriate disinfectants (Appendix E). Understanding what kills causative agents will help you to devise and apply effective control measures.

3. **Maintenance**. Understanding how a disease agent is maintained within a host population is vital for devising effective control and eradication measures. For example, FMD virus might be maintained in carrier animals or sub-clinically affected animals. While these may or may not pose a risk to susceptible animals, it is important to be aware of the potential for FMD-infected animals to be present without showing clinical signs. This can be important when designing a control programme. A control programme that targets only animals showing clinical signs will not control sub-clinically affected animals or animals incubating disease. Therefore, these animals may continue to pose a threat to susceptible animals and facilitate continued spread of disease. By understanding the risk presented by these animals, control measures can be expanded to include all animals that may have been in contact with infected animals, regardless of whether they are showing clinical signs.

6.2 Implementation of control measures and biosecurity

- STOP animal movements — provide instructions to the farmer about how to manage stock without moving them.

- Leave vehicles outside the farm when you make a visit.

- Wash and disinfect all equipment and vehicles before leaving farms.

- All people leaving a farm must disinfect footwear and change clothes.

- Nobody should leave an infected farm to visit other livestock premises.

- Minimise the number of people and vehicles entering/leaving the infected premises.

Implementation of emergency control measures and biosecurity should be rapid and based on concepts of preventing transmission of infection between infected and susceptible animals. Control measures should be maintained until disease has been eradicated and should be

supported by strong communication and public awareness campaigns. If farmers, the public and other stakeholders are not well informed of the reason for implementing disease control measures and the importance of maintaining these measures, it is likely that there will be poor compliance. It is also important that, as a veterinarian or veterinary para-professional, you set an example for others to follow by using good biosecurity procedures during your work in an affected area.

Communication and control measures

You should also discuss the control measures to be implemented with stakeholder groups. Stakeholders will include anyone affected by the outbreak or by the control measures imposed. Stakeholders will include local authorities, police, municipal officers, livestock traders, livestock-market owners, other farmers and the general public. How information is disseminated to these stakeholders will depend on the local situation. Group meetings in an affected village or area are often useful ways of providing information while also providing an opportunity for stakeholders to raise questions or concerns about how disease is being managed. Similarly, village meetings should be organised in the villages that are neighbouring those that are affected to provide advice on biosecurity and preventative measures. For example, if a village (Village 1) is infected with FMD and the neighbouring village (Village 2) is free of disease and both villages usually share a water source on the main road, the farmers from Village 1 should be told not to take animals to the water source, because they risk spreading infection from that village to other areas. It is also important, however, to advise farmers from Village 2 to avoid taking animals to that water source while the outbreak is occurring, as this will prevent their animals becoming infected and taking the disease back to their village. Common water sources are an area where there is often mixing of animals and therefore increased risk of disease transmission.

Practical implementation of control measures

All movement of susceptible livestock should be stopped in affected and adjacent areas. This includes any movements that allow contact between infected and susceptible animals and includes movement for trade as well as normal daily movement for work or grazing. Animals that are not showing clinical signs but have come into contact with affected animals should be included in movement controls. Whenever more cases of disease are found, the control area should be expanded to include the village tract and surrounding areas in which susceptible animals may have grazed. Animal movement controls should remain in place until the outbreak has been brought under control. Controls

should only be lifted 30 days after clinical signs in animals in the last infected premises have resolved.

Where possible, farmers should be provided with suitable disinfectants and given thorough instructions on how to use them (including how often to change disinfectant solutions and how to get more disinfectant when existing supplies are running low).

Farmers and other livestock keepers/handlers in an affected area should be advised against travelling to other areas where livestock are present and should minimise movement of people and vehicles from the infected/suspect area to other areas. It is not practical or ethical to stop movement of people, but they should be requested to carry out disinfection procedures before leaving the affected area, and others should be told to avoid visiting affected areas where possible. Again, communication is vital and it is important to inform farmers of how easily, and by what means, infection can spread. If farmers and other stakeholders have a clear understanding of why the control measures are necessary, they will be more likely to comply with your directions.

6.3 Treatment of affected livestock

The type of treatment required for affected animals in an outbreak will depend on the particular disease involved and on the treatments available to you. Some specific instructions for the treatment of FMD and CSF are provided below. It is important to note that there is no specific treatment for viral diseases (excluding anti-viral drugs, which are not readily available or cost-effective for domestic livestock). Supportive treatment should be provided during the acute stages of disease. Where sound scientific justification exists, antibiotics may be used in very select cases to treat existing secondary bacterial infections. However, use of antibiotics to prevent secondary infections, or without sound scientific justification, is not recommended.

When attending an infected area to treat animals, you must follow strict biosecurity measures. Veterinarians have been known to cause the spread of disease from infected areas to non-infected areas following visits to treat infected animals.

Foot-and-mouth disease

The following measures should be taken for livestock affected with FMD. Some of the treatments described may not be available to you or may not be affordable by the

farmer, while others require minimal materials and should be available to all. While every situation is different, the information provided below should provide guidance when you are faced with the need to treat animals involved in an outbreak of FMD.

1. **Responsible use of broad-spectrum antibiotics**. While FMD is a viral disease and will not, therefore, be responsive to antibiotics, secondary bacterial infections of lesions may occur. Where such secondary infections become debilitating, treatment with appropriately selected antibiotics may be warranted. However, irresponsible use of antibiotics risks development of antibiotic resistance, which is an increasingly serious issue globally. Use of antibiotics prophylactically to prevent secondary bacterial infections, or without sound scientific justification, is irresponsible and not recommended.

2. **Advise the farmer to provide soft bedding and soft feed**. This will make the animal more comfortable and encourage eating when lesions in both the feet and mouth are severe.

3. **Extra attention should be given to suckling young in case they are prevented from suckling due to maternal teat lesions**. Additional milk should be made available to ensure that the young are adequately fed during this time. Milk should be taken from the same species of animal as the young. Young animals should not be taken to another village or farm to be fed as this may spread disease.

4. **Affected animals should have easy access to water without walking**, particularly when lameness is severe.

Classical swine fever

There is no treatment for CSF. Affected animals should be slaughtered for welfare and for control purposes. The disease should be explained fully to the farmer so that he/she understands the seriousness of the disease and the need for such severe control measures. The clinical signs of CSF vary from sub-clinical (and therefore unapparent infection) to severe disease and death. Livestock owners are likely to reject the request to slaughter animals when sub-clinical infection is present and animals appear healthy. Good communication is essential to maintain the trust and support of livestock owners in this situation and to help them to understand the seriousness of the disease and the need to cull infected animals.

7. Tracing cases

Every outbreak of an infectious disease will have a source of infection, and in almost all situations, there will be spread of disease from this initial source. The source of infection can be anything capable of bringing a disease agent from an infected animal in one area to a susceptible population in another area, or at another time, and establishing an outbreak. Possible sources of an outbreak might include:

– An infected animal showing clinical signs that is introduced to a new area with susceptible animals

– A sub-clinically infected animal, or an animal incubating the disease, that is introduced to an area with susceptible animals

– A vehicle or person that has visited an infected area and carries the disease agent to an area where there are susceptible animals

– Contaminated meat from an infected animal, where this meat is consumed by susceptible animals

The spread of an outbreak involves the transmission of the disease to other animals and other areas. The spread of disease from an initial source can be by:

– Movement of infected animals to areas where there are susceptible animals

– Direct contact between infected and susceptible animals that share common areas

– Movement of contaminated people, vehicles or equipment from an infected area to other areas where there are susceptible animals

– Any other means of transmitting the disease agent from an infected animal to a group of susceptible animals

In order to find the source of an outbreak and trace its spread, it is important to understand what materials (animal, human, vehicle, and equipment) might be able to transport the disease agent between infected and susceptible animals, and also the types of infected animal that can directly transmit the disease agent to susceptible animals. All of these potential methods of carrying disease from one infected animal (or area) to an animal (or area) free of disease are known as risk materials. A description of the transmission dynamics and a list of risk materials for FMD and CSF are provided in Appendices B and C.

The method of identifying the source of an outbreak and finding the spread of that outbreak is called **tracing**. Tracing the source of an outbreak is known as 'trace-back'; tracing the spread of an outbreak from an infected source is known as 'trace-forward'. The information collected during tracing can be used to:

– Find affected areas and implement control measures in those areas to limit further spread

– Find the source and implement control measures to prevent further exposure to the source

– Provide information on possible sources and routes of spread to help prevent future outbreaks

7.1 Defining an outbreak

In order to understand and identify the source and spread of an outbreak, it is important to determine when one outbreak ends and another begins, as each individual outbreak will have a different source and must be traced separately. To determine what constitutes a single outbreak, and what constitutes a separate outbreak, we must define exactly what we mean by the term 'outbreak'. To do this, we use an outbreak definition. The outbreak definition below is used by the SEACFMD campaign to define what is meant by a single outbreak of FMD.

> An FMD 'outbreak' is defined as the occurrence of FMD in one or more animals on a farm, in a village, or in a group of animals sharing a common area (e.g. pastureland, watering point, slaughterhouse, market). All cases occurring within 2 weeks of the previous case are regarded as part of the same outbreak.

You can see that the definition provided above explicitly takes into account the three elements of animal, time and place. The space aspect describes how affected animals that belong to the same farm, village or common area (pastureland, watering point, slaughterhouse, market) are considered to be part of the same outbreak. This accounts for local transmission between animals that come into regular contact and share common areas. For example, if two villages use a single grazing area where animals mix and FMD is found to be present in both villages, this will constitute a single outbreak and will be assumed to have originated from a single source.

The time aspect is also important when defining an outbreak. According to the definition above, to be considered part of the same outbreak, additional cases must occur not longer than two weeks after a previous case. The two-week time period relates to the maximum incubation period of FMD,

so that animals that are affected more than 2 weeks after previous cases are unlikely to have been directly infected by the previous cases and must have been infected by a different source. The later cases are then said to belong to a new outbreak.

For every separate outbreak, tracing should be carried out to determine the source and spread of disease. This allows for control measures to be implemented in order to prevent continued exposure of susceptible animals to the source of disease, and to prevent further spread of the disease from infected to susceptible animals.

7.2 Tracing windows

Before you can begin to trace the source and the spread of an outbreak, you will need to gather information to construct a 'tracing window'. A tracing window refers to the most likely period of time during which the disease could have been introduced to an area (tracing window for source), or the most likely period of time during which the disease may have spread to another area (tracing window for spread).

The tracing window for source is used to guide further questioning on the trace-back of an outbreak (to find the source), and the tracing window for spread is used to trace-forward (to find the spread) of an outbreak. We will consider here the information required for construction of a tracing window and how to construct the tracing window based on this information. We will then consider how to use the tracing window to guide questioning to find the actual source of disease and identify potential areas to which the disease could have spread. Table 2 outlines the information that will need to be collected to construct a tracing window.

The information that is collected from the field and from other information sources is used to determine the main parameters needed to construct a tracing window. These are:

1. The minimum and maximum incubation periods of the disease

2. The identity of the index case farm

3. The date clinical signs started on the index case farm

Incubation period is defined as the period of time between the date of infection and the date of onset of clinical signs. The incubation period for any disease will be given as a range, e.g. the incubation period of FMD is 2 – 14 days.

The index case (animal, household, farm or village) is the first case (animal, household, farm or village) identified as infected during an outbreak. If we are investigating an outbreak of disease in a village and farms are the epidemiological unit of interest, the index case can be identified by interviewing farmers. A group of farmers might be asked which farm in the village was first affected by the disease, or which farmer first noticed disease in his/her animals. Alternatively, you could approach each farmer individually and ask them when they first noticed signs of disease in their animals. Identifying the index case is important for tracing, as the index case is assumed to be the case which was exposed to the actual source of the outbreak.

The information collected during the process of outbreak verification (and described in Chapter 5) should be sufficient to construct tracing windows. This information will usually be collected using the outbreak investigation form. In this chapter, we will continue with the same example introduced in Chapter 5, in which it was shown how to make a diagram of an FMD-infected village, including details of the time and place of FMD-affected farms. It will be shown here how we can use this information to construct a tracing window.

We can see from Figure 13 that FMD was first recognised in Farm 1 on 12 October 2008. Therefore, for this example, we would say that the index case was Farm 1 and we assume that the other farms were infected as a result of spread of disease from Farm 1 through sharing of grazing land, visiting common water sources, and other means of transmission between infected and susceptible animals. Based on the clinical signs observed in affected animals, you suspect that the disease responsible for this outbreak is FMD. You then look up the incubation period for FMD and find it to range from 2 to 14 days (Appendix B).

Table 2: Information needed to construct a tracing window

Information required	Information source
Details of the disease cases (affected animals, farms, villages etc.) in area where investigation is being conducted.	Field investigation: outbreak investigation form; village meeting; individual questioning.
Date when clinical signs first seen for each case (animal, farm, village etc.).	Field investigation: outbreak investigation form (Appendix A); village meeting; individual questioning.
Minimum and maximum incubation periods for the suspected disease.	Text books, OIE Terrestrial Animal Health Code, Appendices B and C of this manual.
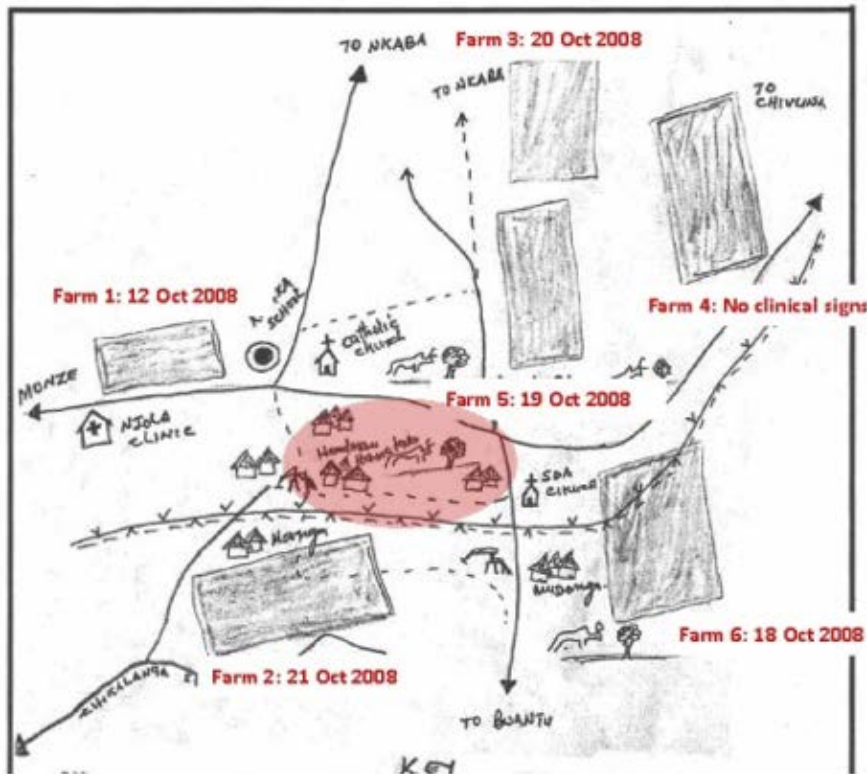
Figure 13: An example of recording cases in time and place.

We now have the following information that will allow us to construct a tracing window:

1. The incubation period of FMD is 2 to 14 days

2. The index case is Farm 1

3. The date of onset of clinical signs on the index case farm was 12 October 2008

Constructing the tracing window for the infection source

When constructing the tracing window, we will refer to the day when clinical signs are first seen on the index case farm as day 0. So, for our example, day 0 is 12 October 2008. Once you have identified day 0 you will need to calculate the time period during which the index case must have become infected such that it would start to show clinical signs on day 0. By doing this, you are identifying when the index case could have been exposed to the source of disease. This time period is calculated using the incubation period. The incubation period is used because this represents the time from infection to onset of clinical signs and therefore accounts for the delay between exposure of the index case

to the source of disease and onset of clinical signs in the index case.

To calculate the earliest time at which the disease could have been introduced to the index case, the maximum incubation period is subtracted from day 0. So, from the example, we would say that the earliest time that disease is introduced to Farm 1 is 12 October minus 14 days (the maximum incubation period for FMD). So, the earliest time infection could have been introduced is the 29 September 2008. We say, therefore, that on 29 September 2008 the tracing window opens.

We then need to know what is the latest possible time that the disease could have been introduced for clinical signs to be seen on day 0. To calculate this, we subtract the minimum incubation period from day 0. For our example, we subtract 2 days (minimum incubation period for FMD) from 12 October 2008 (day 0). So, the latest time that infection could have been introduced is the 10 October 2008. We say that on the 10 October 2008 the tracing window closes. The construction of a tracing window for the infection source is shown in Figure 14.
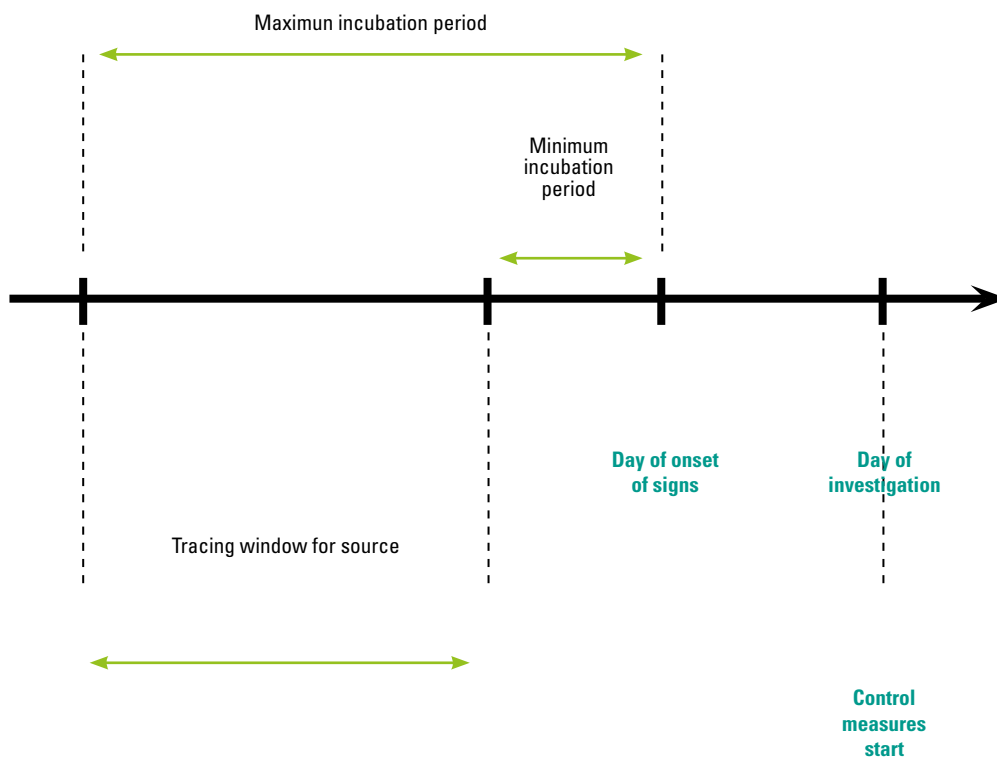
Figure 14: Construction of a tracing window for the infection source.

Using the tracing window for the infection source: trace-back

As described earlier, the purpose of tracing is to identify the source of an outbreak and target control measures at limiting further exposure of susceptible animals to that source. Once the tracing window has been calculated, this should guide further questioning on risk materials being introduced to the area during that time, which could represent the source of infection. In this context the term 'risk materials' refers to anything on which a disease agent can be transferred from an infected animal to a susceptible animal. Risk materials for FMD and CSF are listed in Appendices B and C.

Using our example, we have identified the index case as Farm 1 and the tracing window for source as 29 September 2008 to 10 October 2008. Therefore, we need to question the farmer from Farm 1, and also other farmers and stakeholders, regarding possible risk materials that were brought onto Farm 1, or which may have come into contact with animals from Farm 1, between 29 September and 10 October 2008. This might include purchase of an infected

animal (this animal may not have been showing signs of disease when it arrived on the farm, and may not even have developed clinical signs of disease), delivery of contaminated animal feed, a visit from a person living in an area where there is an outbreak of FMD, or a visit by a veterinarian or village animal health worker that has previously treated infected animals and not followed correct biosecurity procedures. The farmer must be thoroughly questioned about all possible routes of infection onto the farm. Each potential source that is identified should be followed up by the investigating team to determine whether or not it could represent the source of disease.

In this example, we might establish that, between 29 September and 10 October, Farm 1 had bought draught cattle from a livestock market in another township and had been visited by someone from a village nearby that keeps cattle and pigs. We might also find out that the manager of Farm 1 does not buy feed from other farms but grows his own animal feed. You must follow up both the purchase of the cattle and the visit from the nearby village. On following up, you find that there has been no reported outbreak of disease at the livestock market where the cattle

were purchased, or at the village of the person who sold the cattle. You then follow up the person that visited and found that his animals had, indeed, been showing signs of disease before he visited Farm 1, and he described clinical signs similar to FMD. This is the most likely source of disease and control measures should be implemented on that person's farm to prevent further exposure to the source.

Constructing the tracing window for spread

To identify where a disease may have spread, another tracing window must be calculated — the tracing window for spread. The tracing window for spread covers the period of time during which infection could have been transmitted from the outbreak area which you are investigating, to other areas. It is important to identify the areas to which disease may have spread so that control measures can be implemented in those places to limit further, ongoing spread of disease. The tracing window for spread extends from the earliest time that the disease could have spread from the infected area up until the time that no further spread is possible. The earliest time the disease could begin to spread is taken to be the same as when the infection is introduced to the area. So, the tracing window for spread opens at the same time as the tracing window for the source. To calculate the earliest time when disease could have spread from the outbreak area, subtract the maximum incubation period from day 0.

From our earlier example, day 0 was 12 October 2008. Therefore, to calculate when the tracing window for spread opens, subtract 14 days (the maximum incubation for FMD) from 12 October 2008. This means that on 29 September 2008 the tracing window for spread opens. The tracing

window for spread closes when no further spread from the outbreak area is possible. The point at which this situation is reached varies considerably in different situations. For example, if no control measures are implemented, there is potential for the disease to spread until all the affected animals have recovered and have stopped shedding virus. For FMD, this is often taken to be at least 30 days after the affected animals have recovered from the disease. If effective control measures are implemented, however, the tracing window may close much earlier, providing the control measures prevent further spread of infection.

Using our example, suppose that the outbreak at the village is reported to you on 21 October 2008 and, when you go there, you discover from your interviews that the earliest case was on Farm 1 on 12 October 2008. During your initial visit on 21 October, you perform a clinical examination of the animals and find that they are displaying symptoms typical of FMD. Based on your suspicion, you decide to implement emergency control measures (see Chapter 6) and you stop animal movement into and out of the village and provide instructions to the farmers and traders in the village to use strict disinfection procedures if they are leaving the affected area. These control measures are implemented immediately and prevent further spread of the disease from the outbreak area. So, we assume that no further spread occurs after 21 October. This, therefore, is when the tracing window for spread closes. Your investigation will then need to continue examining possible routes of spread from the affected area to other areas between 29 September 2008 (when the tracing window for spread opens) and 21 October 2008 (when the tracing window for spread closes). Figure 15 shows an example of the tracing window for spread.
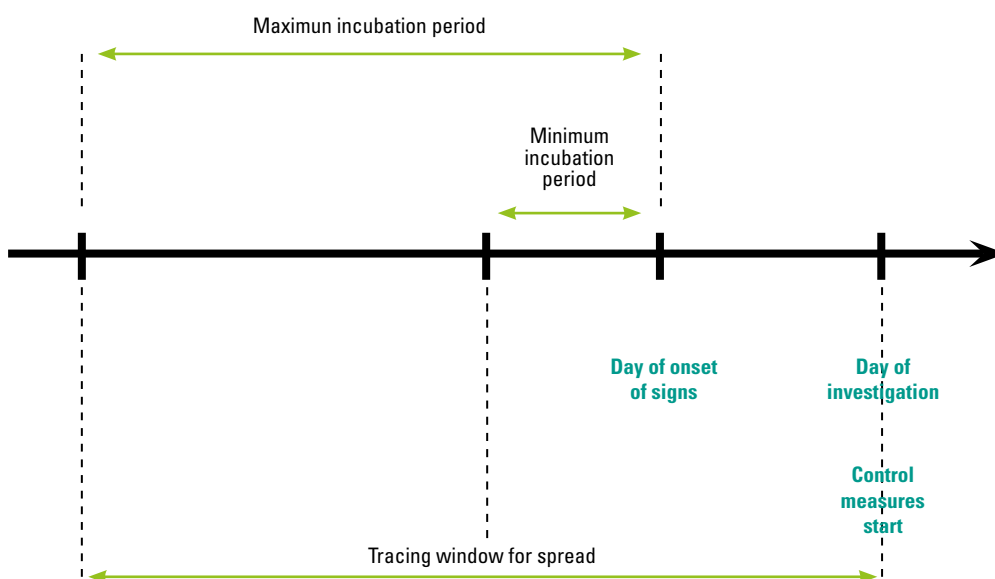
Figure 15: Diagram to show the construction of a tracing window for spread.

Using the tracing window for spread: trace-forward

Once the tracing window for spread has been determined it can be used to guide your questioning on possible spread of disease from the investigation area. This is important as there will often be some spread of disease from the initial source. The areas to which the disease has spread must be identified as soon as possible so that the outbreak can be controlled rapidly. To do this, it will be necessary to ask farmers and other people in the affected area about the movement of risk materials from the affected area to other areas during the period of time identified as the tracing window for spread.

Returning to our earlier example, the tracing window for spread was taken to be from the 29 September to 21 October. You should therefore ask about any movements of animals (even if they did not show clinical signs), people, vehicles, milk or feed from the affected village to other areas during this period. For each area identified as a recipient of potentially infected material, you must follow up to determine whether disease transmission actually occurred. Each area identified should be visited (preferably by an investigation team that has not been involved in the affected

area in order to limit the risk of transmitting disease with the investigation team). Where disease is found, control measures should be implemented immediately and tracing should commence as for the previous affected area. This process of tracing, follow up and implementation of control measures should continue until no further cases are found and the outbreak is eradicated. Remember that if a risk material has been moved to an area recently (within the maximum incubation period of the suspected disease) the animals in that area may be infected but not yet showing clinical signs. Therefore, control measures should be implemented where there is a risk that disease may have spread to an area, and the animals must be kept under observation. Once they have been under observation for at least the maximum incubation period (14 days for FMD) and no clinical signs have appeared, control measures can be lifted. If clinical signs do appear, then control measures should be maintained. A summary of the tracing window methodology for source and spread are shown in Figure 16.
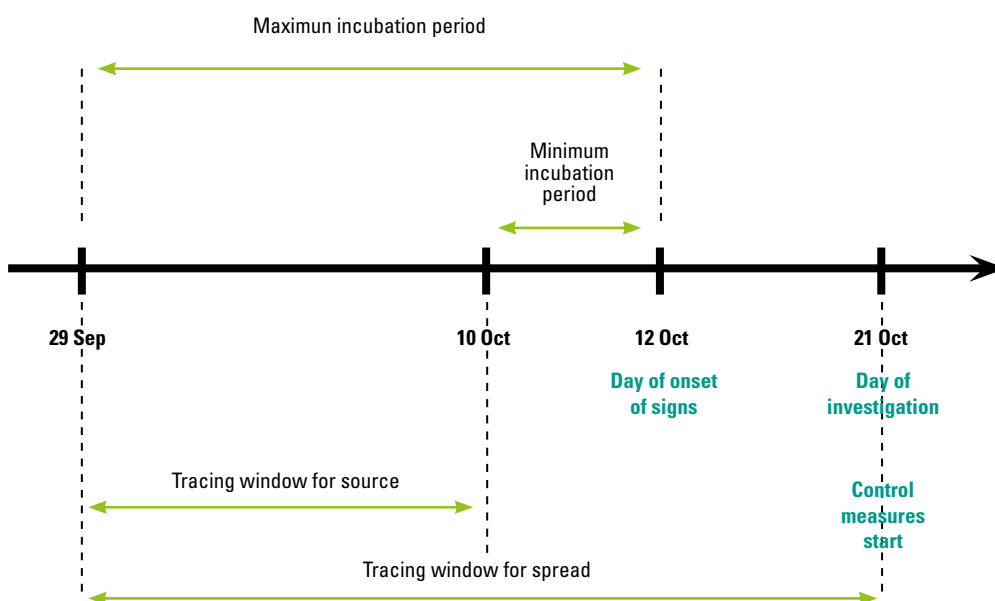
Figure 16: Diagram showing construction of tracing windows for source and for spread using the example of an FMD infected village.

8. Collecting samples

Collection of samples for laboratory testing is an important part of the investigation and can be used to confirm a diagnosis, determine the strain of the agent involved and, in some situations, provide additional information on the epidemiology of the disease. Consideration of the type of samples to collect and how they should be stored and transported to the laboratory are important, because these decisions will influence the diagnostic value obtained from the samples when they are examined (tested) in the laboratory.

Details of sample collection, transportation and submission are provided in Appendix F.

9. Participatory epidemiology

Participatory epidemiology (PE) is the systematic use of participatory approaches and methods to improve understanding of the patterns of diseases in populations and thereby improve options for animal disease control. PE developed from participatory rural appraisal (PRA) in the 1980s, when veterinarians started using participatory methods in community-based projects in Africa and Asia. Both PE and PRA are multidisciplinary approaches to various development problems in rural communities, but PE evolved with a focus on livestock diseases (Catley *et al.*, 2012).

The term 'participatory' is used to refer to the essential involvement of communities in defining and prioritising veterinary-related problems, and in developing solutions to veterinary service delivery, disease control or surveillance. PE is based on conventional epidemiological concepts and allows for the investigation of interactions between host, agent, and environment, but in a social context of disease transmission. It is based on what is called 'existing medical knowledge' (Ameri et al., 2011).

In the context of infectious disease outbreak investigation, the purpose of PE is to get a whole community to take part in achieving consensus about what has happened in the past (e.g. how disease was introduced) and what to do in the future (e.g. how best to manage disease outbreaks). PE can also help in documenting the principal lessons learned from recent outbreaks, which can then be used as tools to inform veterinary preparedness and response plans for future outbreaks. In Section 5.1 we briefly described how to collect useful information from stakeholders, in the next paragraphs we will discuss how PE can further help to refine data collection and offer new insights into the investigation.

9.1 Participatory methods

The active participation of communities in analysing and prioritizing local disease problems, and their involvement in the design and implementation of research, surveillance, or disease control activities, is critical to achieving relevant and sustained benefits. At the initial stage of a participatory process, the relationship between outbreak investigator and community members should be based on a common understanding of the objectives of the process, and the emphasis should be on joint analysis (Catley et al., 2012).

Participatory approaches are based on open communication and transfer of knowledge, using a toolkit of methods guided by some key concepts and attitudes (Jost et al.,

2007). The key principles of participatory appraisal that the investigator needs to keep in mind when collecting information are listed in Appendix G.

Four main participatory methods are used in PE (Table 3) (Jost et al., 2007; Catley et al., 2012):

1. Informal interviews
2. Focus groups discussion
3. Visualisation methods
4. Ranking and scoring

A basic assumption is that investigators cannot fully anticipate the priorities and problems of the community they study. Thus, the participatory process empowers the stakeholders, since they are the ones who identify and describe the problems. This ensures that field approaches are flexible and allow time for the 'discovery' of new information.

Information gathered using participatory methods must always to be complemented by information from other sources, such as:

– Secondary information sources: obtained before going to the study area and as the study is conducted

– Direct observation: people, animals, housing, environment, etc. while in the study area

– Laboratory diagnostics: field diagnostic tests complemented by sample collection and analysis by a regional or national laboratory for confirmation

All information collected is then validated by crosschecking, using multiple techniques and informants: a process called 'triangulation' (Fig. 17). In PE, two types of methodological triangulation have been of particular relevance: 'within-method' and 'across-method' triangulation. Within-method triangulation can be explained using the example of an interview during which the researcher crosschecks information provided by an informant during the interview itself (also known as 'probing'). Across-method triangulation uses two or more different methods to study the same research question. For example, when used for disease investigation or exploratory studies, triangulation can be carried out by crosschecking information within specific participatory methods, by comparing the findings of different participatory methods, and by comparing findings of participatory and conventional veterinary diagnostic methods.

Table 3. Types of veterinary information collected using participatory epidemiology methods.

Method	Information
1. Informal interviews	
Semi-structured interviews (SSI)	Used in most PE studies and in combination with visualisation, and ranking and scoring methods; also used as a stand-alone method
Time-line	History and timing of disease events
2. Focus groups	
3. Visualisation methods	
Participatory mapping	Livestock movements with respect to the location of grazing areas and water points and spatial exposure to disease vectors
Seasonal calendars	Seasonal variation in disease incidence; seasonal variation in human livelihoods; seasonal variation in the livestock trade and in the consumption of livestock products; seasonal variation in contact with disease vectors, neighbouring livestock and wildlife; seasonal variation in vector populations
Proportional piling	Age structure of livestock herds; disease incidence and mortality estimates by age group; impact of vaccination on livestock mortality; case fatality rates
Radar diagrams	Analysis of disease control strategies
4. Ranking and scoring	
Simple ranking	Analysis of disease control strategies
Simple scoring	Prioritisation of livestock diseases
Matrix ranking	Analysis of disease control options
Matrix scoring	Local characterisation of the clinical signs and causes of disease; local characterisation of disease vectors; comparison of clinical diagnoses of livestock keepers and veterinarians; analysis of veterinary service providers
Before-and-after scoring	Impact of veterinary services on livelihoods; impact of diseases
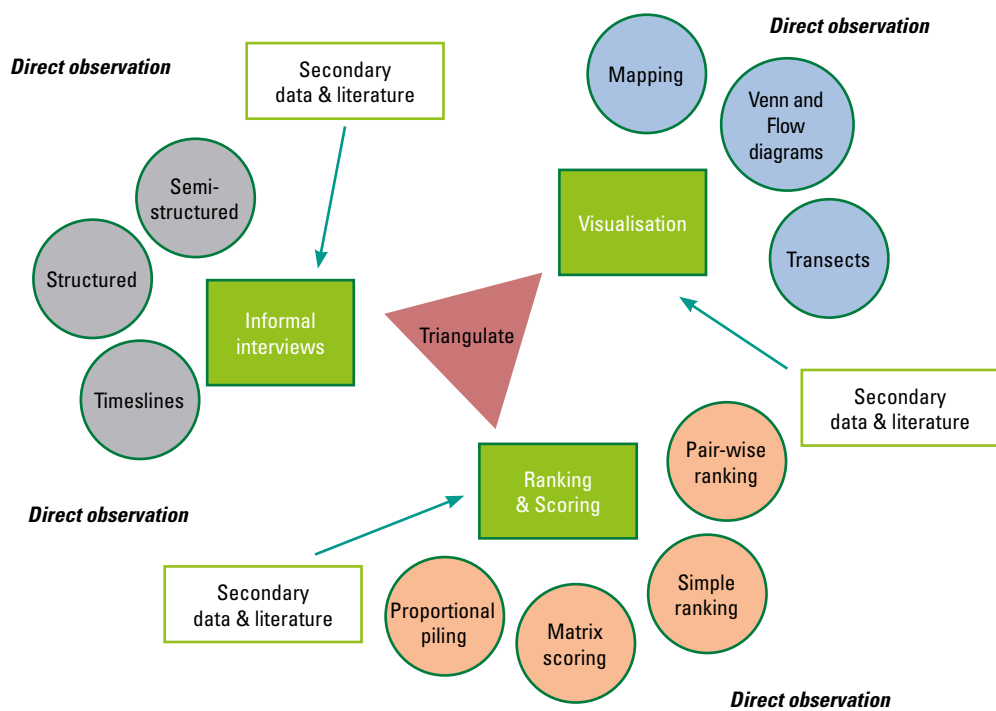
Figure 17. Types of veterinary information collected using participatory epidemiology methods (Catley, 2005)

9.2 Attitudes and behaviours in PE

An important aspect of participatory approaches is the way we interact with other people. This interaction determines the relationship and trust that develops between investigators and local people, and affects the types of issues and information that people are willing to discuss in an open manner. If we look at this issue from an epidemiological perspective, the relationship between investigators and livestock keepers is a key factor affecting the reliability and validity of data. If informants are concerned that investigators have a 'hidden agenda', will use the information solely for selfish purposes, or may pass information to authorities, then their participation will be poor. In addition, if informants consider outsiders rude or arrogant, or only interested in their own opinions, the discussion will not be very constructive.

For the interaction to be meaningful, investigators must believe that an informant has something useful to say. This means respecting local views and opinions, and being open to ideas that may not necessarily agree with modern science. This does not mean that, as veterinarians, we must automatically accept all indigenous knowledge as valid and useful. The idea is to identify local knowledge and skills that seem to agree with our professional expertise, and explore this local knowledge.

9.3 Team work

Many PE methods work best when a team of two or more investigators work together. Within the team, roles should be clearly defined.

- One person should be the facilitator. The facilitator introduces the session, asks questions, explains the method, and checks the information as it arises from the informants. Therefore, the facilitator interacts directly with the informants and does not need to write anything down during the session. In other words, the communication flow is not interrupted because the facilitator keeps stopping the discussion in order to write down what has been said.

- Another team member acts as the recorder. This person usually sits slightly back from the group and records the discussion or results of scoring methods as they arise. The recorder also watches the group dynamics and looks to see who talks in the group and who does not. If necessary, the recorder can remind the facilitator

to include people who are not contributing to the discussion.

The team members need to carefully prepare how they are going to run each session and who is going to say what. It can be very confusing for informants if, for example, the team members interrupt or contradict each other.

9.4 Examples of useful PE methods for outbreak investigation

An important aspect of PE methods is their capacity to reach illiterate people and involve them in description and analysis of local problems. With methods requiring people to either write or understand text, illiterate people can easily become isolated and may not contribute because they are embarrassed, or because literate people dominate the discussion. Many PE methods, such as interviews, matrix scoring, mapping, seasonal calendars, and proportional piling can be conducted using no written words. With these methods, disease signs (such as mouth ulcers and feet lesions for FMD, and skin lesions for CSF), disease causes, parasites, livestock types and other items can be represented by either everyday objects or pictures. Pictures can be drawn or printed on to pieces of card and these cards form the 'labels' for the method.

Semi-structured interviews

An interview is a focused conversation between two or more people. It is a method of collecting data by talking to people and asking questions. In structured interviews, the instrument used to collect data is a questionnaire. Questionnaires often use closed questions, which can usually be answered with 'yes' or 'no' or a short response. Generally, questions are asked in a manner such that the answers fall within an expected range of answers. By restricting an interview in such a way, one risks losing valuable information, viewpoints, and the context behind a response. Remember, if it appears that a response is not rational, then we have failed to understand some key factor in the situation. By avoiding closed-ended questions, we provide the respondent with the opportunity to explain to us the rationality behind a response.

For this reason, interviews in PE are semi-structured and the interviewer uses checklists of topics to be covered rather than a structured questionnaire. The interviewer introduces a topic using an open-ended question, i.e. a question

designed to encourage a full, meaningful answer using the responder's own knowledge and feelings. Typically, these questions begin with why, when, how, what, where, who? After listening to a response, the interviewer can probe further with clarifying questions.

An example of an open-ended question would be: 'What diseases affect your cattle?' This allows the respondents to provide direction to the interview and describe problems in their own terms. Once the participants have noted and described problems, the team can then ask probing questions to fill in any gaps and to check for internal consistency within the individual accounts. Summary guidelines for semi-structured interviews are listed in Appendix G.

Focus groups

Holding a focus group discussion (FGD) is a method of collecting qualitative data which is expressed in words, not numbers, and there is no numerical generalisation. The purpose of FGD is to obtain in-depth information on the concepts, perceptions and ideas of a group. This component involves general discussion using open-ended questions to explore farmers' understanding of animal health services in their area and their knowledge of public health, diseases affecting their livestock, and other diseases, including zoonoses (Sihavong, 2009). FGD are often used to focus research, for example, when broad research ideas need to be refined or if there is the need to formulate appropriate questions for structured (quantitative) studies. They can produce a lot of information quickly and they are very useful for exploring beliefs, attitudes, behaviour and concepts in a population. They can assist in understanding problems and results from interventions and explore controversial topics.

Figure 18. A Dutaik (traditional Burmese meeting approach) meeting in a village, Myanmar. (Oo, 2010)

Specific groups are targeted based on the type of information needed (e.g. within the village, a group of farmers in charge of livestock). This group can then be divided into sub-

groups with particular characteristics, as this will allow more detailed information to be collected (e.g. within the group of livestock farmers, the ones that are responsible for trading). FGD vary in duration, but typically they are 1 – 1.5 hours long; often, the first discussion of each sub-group is longer than subsequent discussions. The number of FGD required depends on project needs and different types of groups; generally, at least two groups per sub-group are interviewed. The methodology for running an FGD is described in Appendix G.

Participatory mapping

Mapping is a type of visualisation method, which is a popular participatory tool among animal health workers. Mapping is a useful method for the following reasons:

– Both literate and non-literate people can contribute to the construction of a map (as it is not necessary to have written text on the map)

– When large maps are constructed on the ground, many people can be involved in the process and contribute ideas. People also correct each other, and make sure that the map is accurate

– Maps can represent complex information that would be difficult to describe using text alone

– Maps can act as a focus for discussion

Mapping is best used with a group of informants, say between 5 and 15 people, such as in a Dutaik meeting (Figure 18). Find a clean piece of open ground. Explain that you would like the group to produce a picture showing features such as:

– Geographical boundaries of the community; these boundaries should include the furthest places where people go to graze their animals (Fig. 19)

– Main human settlements

– Roads and main footpaths

– Rivers, wells and other water sources

– Grazing areas (particularly common grazing areas), farmed areas, forests and other natural resources

– Ethnic groups

– Movements of livestock by livestock type

– Spatial contacts with herds from other communities or wildlife

– Outbreak index cases

– Source of infection

– Disease spread

Figure 19. Example of a map of a Northern Laotian village affected by FMD. The map shows the location of the main livestock types, areas of cultivation, geographical features and possible sources of disease. The map was produced by district veterinarians as part of an FMD outbreak investigation training group exercise (OIE, unpublished data)

Figure 20. A 'risk map' showing an HPAI outbreak in an Indonesian village.(Hendrickx and Pissang, 2010)

In order to collect valuable information about a disease outbreak that recently occurred in a neighbourhood, or in a village, participants should be asked to list all of the disease determinants that they think are associated with the outbreak and then draw a map of that outbreak, showing as many of the disease determinants as they can. This type of participatory map is called a 'risk map' (Fig. 20), because the participants produce an image that shows the spatial distribution of disease risk. When the group is happy that the map is finished, they will need to explain the key features of the map. The process of 'interviewing the map' enables investigators to learn more about the map and pursue interesting spatial features. The methodology for developing an outbreak risk map is described in Appendix G.

Proportional piling

Proportional piling is a tool that allows respondents to give relative scores to a number of different items or categories according to one criterion. The scoring is done by asking participants to divide 100 counters (beans, stones or

similar items that are familiar to the community and locally available) into different piles that represent the categories. For example, the community could give scores to a set of disease problems (the categories) according to the impact that the diseases have on their livelihood (the parameter). Alternatively, the community could be asked to score the diseases according to how frequently they occur. Semi-quantitative data is collected by recording the number of counters in each category.

It is extremely important for outbreak investigation, as it can be used to define which risk factors have majorly contributed to the outbreak (Fig. 21), the relative incidence of the disease in the village, or the relative impact of the outbreak on the livelihoods of farmers. An example of how to use proportional piling is described in Appendix G.

Matrix scoring

In participatory epidemiology, clinical case definitions are important tools for probing local medical or veterinary
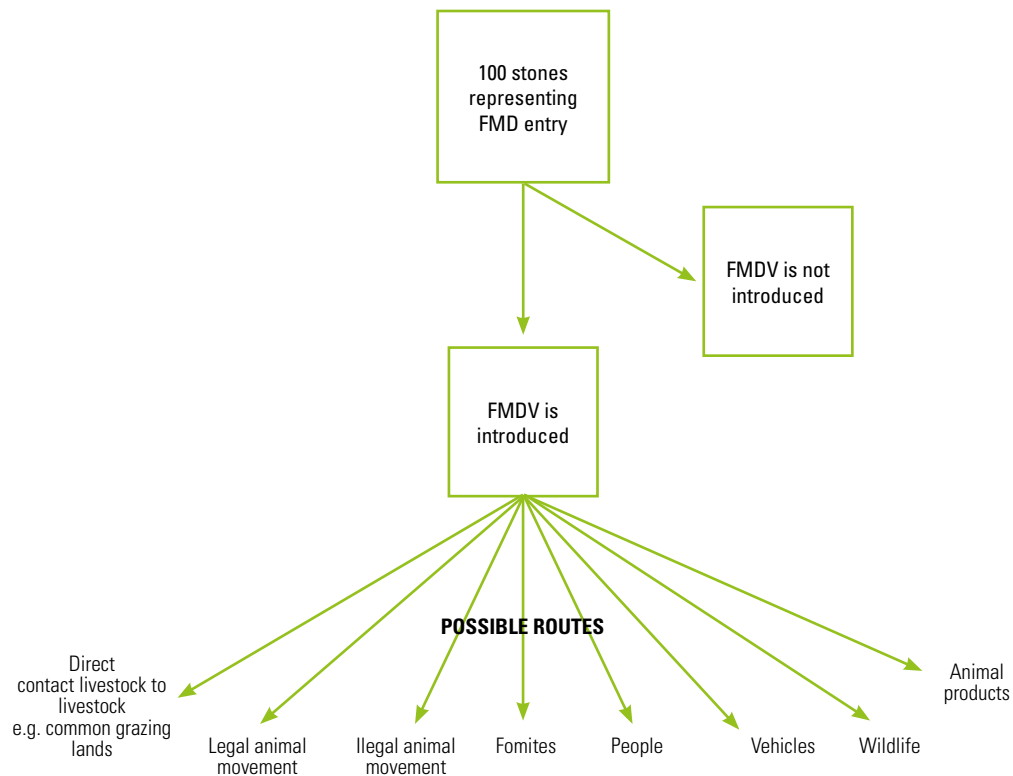
Figure 21. Example of the proportional piling method to estimate the risk pathway of FMD entry. Using a pile of 100 stones to depict an age group, the informant was asked to divide the stones to show the pattern of 'FMDV being introduced during the last year' and 'FMDV not being introduced during the last year'. The pile of stones representing FMDV being introduced was then sub-divided by the informant to show the main transmission routes and the extent to which each of them was responsible for viral introduction.

knowledge, and linking that knowledge to a practitioner's knowledge about diseases and syndromes. We use signs, symptoms, and epidemiological criteria to arrive at a diagnosis. Clinical case definition is particularly important to define the occurrence of an outbreak; it should provide enough criteria so that the practitioner can arrive at the expected level of certainty in his or her diagnosis, while not being too prescriptive so that important cases go undetected.

Matrix scoring can be very useful during outbreak investigations, as it helps to build a clinical case definition and verify if the outbreak has really occurred. The method is essentially a series of proportional piling exercises in two dimensions, where a list of items, such as diseases, is scored against a list of indicators, such as clinical signs.

An important first step in any study is to understand how community members think about and characterise diseases. You may find, for example, that your respondents mention several diseases, each with their own name in the local language, but to you they seem very similar. Matrix scoring

can be a very useful tool for understanding the symptoms and epidemiological characteristics of the different diseases respondents mention, as shown in Figure 22.
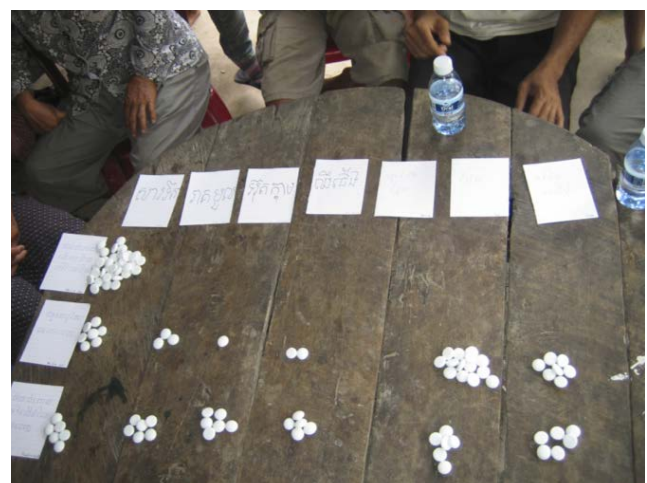
Figure 22. Matrix scoring of cattle diseases against disease signs by villagers in Svay Rieng province, Cambodia. (Bellet *et al.*, 2012)

It is important to note that when a clinical case definition is being used for early detection of outbreaks, it may be quite broad, as this will ensure that it picks up all potential cases. A laboratory test is then required to confirm the diagnosis. The methodology for developing a matrix score for disease identification is described in Appendix G.

Seasonal calendars and timelin es

Many human and animal health problems show seasonal variation. A seasonal calendar can be used to visualise and analyse local perceptions about the seasonality of disease incidence, vector populations, risk factors, farming practices, etc. The seasonal occurrence of diseases is interesting to understand in relation to the seasonality of factors that affect their occurrence, such as climate, management practices, vectors, etc. New or unusual factors may emerge that are important in a particular area. Factors linked to the occurrence of the outbreak can be related to particular seasonal events. In addition, timelines are useful tools for exploring the frequency of key disease events and patterns over time and estimating the duration of outbreaks and other disease events.

Seasons are defined by different characteristics in different regions. Understanding the characteristics that are used to define the seasons in the area under investigation is the first step in creating a seasonal calendar. Other seasonal events (indicators) can then be investigated. Human activities, namely political, religious, and cultural events such as festivals and holidays, can affect movements and disease spread. Other seasonal factors, such as availability of water or presence of vectors, may be of interest, depending on the disease under investigation. Management and marketing practices for livestock may be seasonal due to movements, calving, housing, and buying stock or off-take, and may be significant in terms of zoonoses risk.

Many diseases of interest occur as epidemics at finite time points, or as flare-ups of endemic disease. The interviewer may note the years of major epidemics for various diseases on a timeline. Information on major events, such as droughts and famines or political events may also be included to assist informants in remembering the timing of key disease events. These events may also have an impact on disease occurrence because of the changing movements and habits of animals and people. Their inclusion may allow for triangulation of reported risk factors for disease occurrence. Besides providing information in itself, the timeline will provide a useful reference for triangulating the reports made by the community with information in the official government surveillance system.

During an outbreak investigation, participants can be asked to construct an outbreak timeline. The investigator can decide on a timescale that will capture all of the events during the outbreak, as well as events before and after that had a bearing on the outbreak. Key events that both the investigator and the participants can identify are included (e.g. early warning released). All of the outbreak events are listed and when all the indicators are ready, a timeline can be prepared.

9.5 Data analysis

The flexibility of participatory appraisal allows practitioners to review and analyse data on the spot and make changes to the appraisal checklist. The appraisal team is encouraged to discuss observations as the need or interest arises. Every few days, the team should review the progress of the appraisal in a systematic manner and assess if the right types of questions and exercises are being carried out. Perhaps new elements of the community or a new class of key informants have become known and need to be worked into the interview schedule. Alternatively, a new item for probing or ranking exercises has become a burning issue (Mariner and Paskin, 2000).

It is the spirit of participatory appraisal that analysis is continuous. Hypotheses are continuously refined and focused. The process might be compared to a continuous cycle of the weighing and comparison of factors. Some factors or disease determinants are gradually pushed to the periphery while others are brought into sharper focus at the centre of the enquiry.

The appraisal team can discuss preliminary findings and hypotheses with community members and key informants. These should be advanced as neutral observations, with requests for the respondent's views. Care must be taken not to lead respondents towards endorsement of 'preferred' views. If the team is concerned regarding 'politeness' bias, try presenting the hypothesis in a negative sense. As an example, 'Somebody told us ..., but how can that be true?' If the respondent replies that it is, in fact, true, good support for the hypothesis has been indicated. This subject was also discussed above in reference to probing. Pay close attention to the factors the respondents introduce in considering the findings and hypothesis. This data is more important to the analysis than simple concurrence or disagreement. These factors may suggest new avenues for enquiry and offer further means of triangulation.

A good setting for action-oriented analysis of study findings is a community workshop. These can be formally scheduled with invitations, etc. or more ad hoc. At these workshops, study findings are usually presented in a participatory manner so that the participants debate the findings and, hopefully, reach agreement on their final interpretation.

The outcome of the workshop is a set of agreed points for action, which specify the duties and responsibilities of all parties involved.

10. Communicating your findings to stakeholders

10.1 Preparation of an outbreak investigation report

An important component of an outbreak investigation is the write-up of the investigation report. It is important to document details of the outbreak and the subsequent investigation and response. The outbreak investigation report is a way of communicating information about the outbreak and the effectiveness of your response to your colleagues and superiors within the Veterinary Services. The report will be based mainly on the information gathered during the outbreak investigation, using the standard outbreak investigation form. The following is a description of the general outline of an outbreak investigation report.

1. Title.

2. Basic summary information about the outbreak, including details of location (state-division, district, township, village tract, or village affected), date of onset, date of first visit, species affected (using a table to indicate the total number of animals on the farm, the number affected, and the number dead), and the results of any laboratory tests.

3. A description of the outbreak and how it spread through the village, including details about the animals affected ('animal'), the time of onset ('time') and the location of positive and negative cases ('place') (as described in Chapter 5).

4. A description of your hypotheses about how the outbreak might have started and what were the most likely risk factors/risk materials involved in the introduction of the disease.

5. A description of the emergency control measures and biosecurity measures implemented.

6. A description of future actions that should be taken to fully control the outbreak and also to prevent future recurrence.

Ideally, the outbreak investigation report should be completed as soon as possible after the initial investigation in order to keep all levels of the Veterinary Services well informed. Follow-up reports can be made when new information becomes available (e.g. laboratory results, alteration of control measures, further spread of the initial outbreak).

10.2 Communication in an outbreak

Communication and public awareness is crucial to the success of outbreak investigation and management. The process of communication includes gathering information during an outbreak (organising group meetings, conducting interviews, etc.) and disseminating information and key messages to farmers, the public and other stakeholders. Although communications is placed as the final step in the outbreak investigation process, it should be carried out at each and every step of the outbreak investigation. Notes about communication have been integrated into each of the investigatory steps described in this manual to help guide you during an outbreak.

Setting up village meetings

Village meetings can be very useful for gathering information from and disseminating information to stakeholders.

Organising a group meeting as opposed to conducting individual interviews is often a faster and more efficient way of gathering or disseminating information. The information gathered and shared is often greater in a group meeting, as information can be discussed and cross-referenced amongst the participants. Information on past events is often more reliable, as the participants can help each other remember things. This is referred to as 'group memory' and is often more accurate than individual memory. Group meetings also provide an opportunity to collect and disseminate different sorts of information at the same time.

When holding a group meeting in a village, it is ideal to have all owners of the relevant livestock species present. This is not always possible, but you should aim to include as many of them as possible. The people who attend the meeting should include those who know the animals best - that is, individuals responsible for their daily care. The number of participants at each meeting should be considered carefully: smaller groups are easier to manage, but the 'group memory' tends to be better among larger groups. However, larger groups can be harder to manage and a lot of information can be lost during private conversations between individuals.

In an outbreak, you will generally be organising a meeting without a great deal of prior notice. In this situation, it is important that you arrive as early as possible for the

meeting and are prepared to wait until most people can be there. If possible, you should send a message to the head of the village or local authority as soon as you make a plan to visit the village so that they can organise for the farmers to be gathered at the time of your visit.

Meetings can be held in a range of places, including community halls, schools and temples. They can also be held in the home of a livestock owner, in an open space or under a shady tree.

Participatory approaches to information gathering

Once you have organised the village meeting and the participants are gathered, you can begin to conduct the meeting. This section provides a guide on how to conduct a village meeting using participatory approaches to gather information. Participatory approaches aim to facilitate exchange of information between the disease investigator and livestock owners. An example of participatory methods used in this context in Myanmar is the Dutaik Meeting. Because the Dutaik Meeting is a traditional meeting used for discussion in villages, participants tend to be familiar with the routine and structure of the meeting. As a result, they will usually share information more readily in the Dutaik setting than in other, unfamiliar meeting arrangements.

This section will describe some of the skills and qualities that are needed by a disease investigator in order to conduct village meetings using participatory approaches. It also describes some important methods used in conducting village meetings, such as encouraging participation, listening, building trust, and using particular language.

In order to effectively communicate in an outbreak situation, and when holding a village meeting, the disease investigator should have:

- A good technical knowledge of the disease under investigation

- Fluency in the language used by the participants, if possible (e.g. minority languages or common languages used)

- An ability to speak clearly and an understanding of the culture and social customs within the village

- Knowledge of, and respect for, the skills of the participant livestock owners and a willingness to let them express their opinions

- An ability to encourage participation, particularly of the quiet and shy members of the group

It is not always possible to have all of these skills, particularly if you have recently moved to a new area and you are not familiar with the local area where the outbreak occurs. In some situations, you may be able to use a local contact person to assist you in conducting the meetings. This local contact may have qualities that will assist you to communicate with the participants, such as a detailed knowledge of the local dialect. Note that, if most of the participants are women, it is best to have a woman as the presenter.

All participants should have an opportunity to express their views. There are many reasons why people are reluctant to participate during a meeting. The main one is social status and local tradition and customs. The village head's opinion, for example, may not be challenged by others, or decision-making may be reserved for community elders. In some communities, women or young people may be reluctant to express themselves. You should be aware of all these factors when conducting a meeting, and you should understand social and cultural constraints to participation. A good understanding of the local culture, the village area and the sensitivities of the community will be a great advantage. It is also important to remember that as a veterinarian or veterinary para-professional visiting an area you may intimidate some village members, who may avoid discussing veterinary matters for fear of sounding ignorant. Be aware of this and make them feel comfortable.

At the meeting, be aware of the range of people with different social levels, e.g. the head of the village, government officials, and wealthier livestock owners, etc. Also, remember that livestock owners have their own ranking as well, which depends on their age, their social ranking in the village, the number of livestock they own, etc. To help reduce the social constraints, everyone should sit at the same level (on the ground or on chairs). Sitting in a circle is better than the disease investigator sitting in front of the audience.

Directing specific questions to specific members in a group will encourage participation. If there are some people within the group who do most of the talking, make an effort to repeat questions and aim them at other members of the group.

At the beginning of the meeting you should conduct an 'ice breaking' exercise, particularly if the group members are unfamiliar with you and/or with other members of the group. An 'ice-breaker' aims to get all participants to speak and can be done formally, by asking people to state their names, the number of livestock they have, etc., or informally, by using a game or activity to relax the participants. This will also get them speaking and thinking about livestock.

Effective communication is a two-way process and should not just be a one-way dissemination of information. One of the major roles of veterinarians and veterinary para-professionals is providing advice and information on animal health issues to livestock owners, but it is also important that the veterinary workers listen and take seriously the opinions or observations of livestock owners.

Listening skills are fundamental to rich and meaningful communications. You can listen without actually hearing what is being said because you are too busy interpreting, assuming or preparing a response. Being a good listener means that people who are engaged in conversation with you feel valued, and this gives you a good basis for building a productive relationship. It is easy to have a tendency to interrupt livestock owners to correct misconceptions and provide advice, but you first need to listen and hear what is being said.

Active listening encourages the open communication of ideas and feelings by making the participant feel both heard and understood. Some tips include:

– Look at the person who is speaking —- show that you are both interested in what they are saying and that you understand. If you do not understand, ask questions to clarify.

– Listen to both what is said and how it is said —- pick up on the emotion as well as the words.

– Summarise what you have heard —- show that you have caught the main points.

– Respond politely to all questions, even if they seem naïve.

– Respect the answers and viewpoints that are different from your own; do not belittle learners or other trainers.

Good questioning encourages people to go beyond simply providing information. It prompts them to share their views. Ask open-ended questions rather than closed ones ('yes' or 'no' answers). For example, ask 'What did you do after you saw your animal was sick' rather than 'Did you keep your animal on your premises only after you saw it was sick and suspected FMD'?

It is most effective if you speak the same language and dialect as the group participants. Using the same language and expressions helps reduce the distance between you and the livestock owners and encourages better participation. Your language should be clear and simple and avoid technical terms (something that a trained veterinarian can often find difficult).

You should be aware of local terminology, for example, the disease which you are investigating might be called by a different name in a particular area. This name should be identified at the beginning of the meeting and used when talking to the participants. Make sure that when you

describe, or ask about, clinical signs, that the participants know what you are referring to. Again, some clinical signs may be described using different terminology in different areas. Try to use this terminology when speaking to villagers. When describing the signs of the disease, use pictures and photographs and ask owners to tell you what words they use to describe the signs in the pictures.

By knowing the local setting, the culture, social status, traditions, language spoken, religion, economics, livelihoods, forms of communication and aspirations of participants you will have a much better understanding of the group and be able to relate with them more easily. If you are new in an area and are unfamiliar with the local setting, it is often helpful to have a local contact that can help you and advise you in communicating with the participants.

To be effective in conducting communications during an outbreak, it is important that you build a trusting and understanding relationship with community members. When communicating you need to build, maintain and, if necessary, restore trust. A loss of trust can have severe impacts and can severely limit the effectiveness of your investigation and management of an outbreak. Maintaining trust throughout an outbreak requires transparency (i.e. communication should be honest, easily understood, complete and factually correct).

Confidentiality must be preserved so that people feel comfortable about providing information. This is also important when people report an outbreak. People fear that there could be repercussions if they report the disease, e.g. stamping out without compensation, so they must feel confident and they must trust that you will keep their information confidential.

Disseminating information during an outbreak

During an outbreak there will be important messages that you should give to different members of the community, depending upon how they are involved with the outbreak. This section considers some of the key messages that should be provided during an outbreak, with emphasis on the messages to livestock owners and traders, and on the different groups (or audience) that should be targeted.

Providing clear messages to specific members of the community during an outbreak is vital to the success of managing the outbreak. The key messages that you should be giving to farmers, members of the public and local authorities include:

1. Report. Notify and seek help from a veterinary officer or veterinary para-professional as soon as unusual or

severe signs of disease are seen in your animals. Early reporting of a problem enables rapid detection of serious diseases, such as FMD, before they cause serious social and economic consequences.

2. Stop animal movement. If your animal has, or is suspected of having, disease, it should be isolated immediately from other animals to stop the disease from being spread. When you are notified of an animal disease outbreak in your area, keep your animals on the premises and avoid bringing them to common pasture land or water sources. Movement of animals is a major factor in the spread of livestock diseases. Some disease, such as FMD, can also be spread by animal products, people, vehicles etc., so you should avoid moving animals into or out of an outbreak area.

3. Do not buy or sell sick animals, even if the price is very low. If you do this, FMD will be spread to other areas.

Livestock owners are one of the major audiences during an outbreak, as they are responsible for taking care of livestock and complying with control measures. Livestock owners are also often the people who are most affected by outbreaks of disease in livestock. Livestock traders, dealers and marketers are another essential target group for disseminating information during an outbreak. The messages for traders are the same as those for farmers, but there needs to be an emphasis on the importance of doing the 'right thing' when it comes to buying animals. Animals must be bought from disease-free areas where possible. They should not buy sick animals and should follow regulations for quarantine and disease prevention, where appropriate.

It is not just the livestock owners and traders who are your key audience in an outbreak. You may need to make presentations and organise meetings with the following groups to effectively contain an outbreak. These audiences include:

– Village animal health workers

– Government staff at all levels — commune, district, province

– Border patrol and/or quarantine officers

– Animal inspectors

– Slaughterhouse owners

– Livestock-market owners

– Livestock organisations

– Private veterinarians

11. Case examples: Outbreak investigation

11.1 Outbreak investigation of reported FMD in Maoming, Guangdong province, P.R. China

On 17 February 2013, a suspected FMD case was reported by Donghui Swine Farm in Maoming, Guangdong province in China. The outbreak was reported to have begun on 3 February, but production records suggest that the suspected case may have occurred on 20 January.

The symptoms included a rise in animal temperature, vesicles and ulcerations on the nose and coronets, sloughy hooves, and lameness. Swine showing these symptoms are suspected cases. Blood and lymphoid tissues were sampled from two local slaughterhouses and 97 blood samples were taken from 10 farms within 5 kilometres. All these samples were analysed by the national reference laboratory.

Vesicle on the nose   **Sloughy hoof**

As of 17 February, the attack rate and case fatality rate of breeding swine were 74.6% (88/118) and 0% (0/88), respectively. The mortality rate of piglets was 39.3% (110/280), while 550 fattening pigs were still clinically healthy when culled.

On the affected farm, movement restrictions and disinfection measures were put in place on 17 February. Two days later, on 19 February, all the pigs were culled and safely disposed of by burying. At the same time, within 3 kilometres, transportation of swine, cattle and sheep was forbidden and trading markets were closed.

On 27 February, the suspected cases in Donghui Swine Farm were confirmed as FMD type A. Three blood and eight aetiological samples collected from farms around Donghui Swine Farm were found to be positive. Qualitative analysis shows that the most probable pathway of introduction was a vehicle. The risk of FMD spreading outside the farm is low.

Disinfection   **Deep burial**

11.2 Outbreak investigation of FMDV serotype O Ind2001d in Dak Lak and Dak Nong provinces, Vietnam in 2015

On 10 September 2015, RAHO6 confirmed FMDV serotype O as the virus responsible for an outbreak in pigs in Village No.4, Thanh Nhat commune, Buon Ma Thuot city, Dak Lak province. The pigs were found to be unvaccinated and the first clinical signs, includingpyrexia, salivation, lameness, vesicles and erosions in the mouth and feet were observed on 23 August.

On 30 September 2015, another outbreak occurred in unvaccinated pigs in Village No.9, Tam Thang commune, Cư Jút district, Dak Nong province. RAHO 6 confirmed that the serotype responsible was FMDV serotype O. Clinical signs, including pyrexia, salivation, lameness, and vesicles and erosions in the mouth and on the feet, began on 28 September.
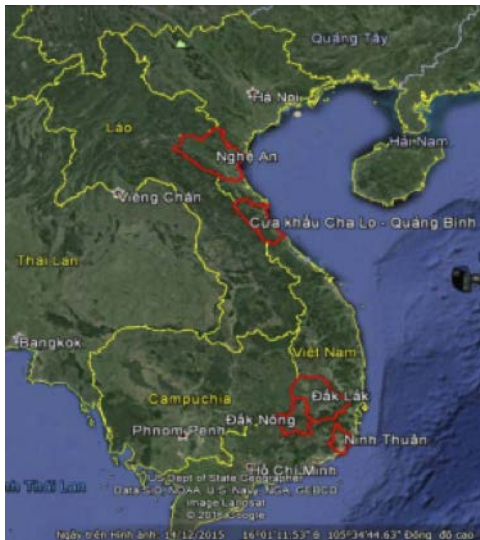
Map showing location of Dak Lak and Dak Nong provinces in Vietnam where FMD serotype O Ind2001d occurred

These outbreaks were not the first outbreaks to be caused by FMDV serotype O Ind2001d. The serotype was first identified in Vietnam on 26 May 2015 following an outbreak in Dak Nong province (> 3 months before these two investigated outbreaks).

The morbidity rate in infected farms was > 90%, while the mortality rate in infected animals was > 25%. Risk of disease spreading comes from animal movements (between and within borders).

Appendices

A. Example outbreak investigation reporting form

Epidemiological Investigation Reporting Form

Reporting Form ID: _____

Investigating Official: _____

Position Held: _____

Signature: _____

Investigation Date: _____/_____/_____

Section 1: Outbreak reporting

1.1 The officer was informed of this outbreak by:

☐ Owner

☐ Livestock volunteer in village or sub-district

☐ Village or sub-district headman

☐ Other *(please specify)*:_____

1.2 Reporting Date: _____/_____/_____

Section 2: Index case

2.1 Name and address of owner of the first case:

_____

i)   Village Name:_____

ii) Subdistrict/District/Province:_____

iii) Coordinates: X _____Y _____

2.2 Date the first signs of disease were noted: _____/_____/_____

2.3 Species of the first case:

☐ Cattle   ☐ Buffalo   ☐ Pigs

☐ Sheep   ☐ Goats   ☐ Poultry

☐ Other *(specify species)*:_____

2.4 If the first case was introduced from another area, please provide details:

i)   Specify date of introduction: _____/_____/_____

ii) Specify source location: _____

2.5 Owner managed diseased animals by _(answer all applicable options)_:

☐ Slaughter _(specify location)_____

☐ Consumption or distribution _(specify location)_____

☐ Carcass disposal (burial or burning) _(specify location)_____

☐ Treatment _(specify treatment)_____

☐ Other _(specify)_____

Section 3: List of cases

Owner Name: Village Name: Village ID Number _(if applicable)_: Subdistrict: District:									
Species	Total number susceptible	Number affected	Number of deaths	Number slaughtered/ destroyed	Date first animal affected	Date last animal affected (if applicable)	Vaccination History		Specify origin if introduced
							Vaccinated date	Vaccinated times	

Section 4: Clinical investigation

4.1 Clinical signs

☐ Fever   ☐ Jaundice

☐ Bleeding/haemorrhage   ☐ Abortion

☐ Drooling saliva   ☐ Blisters on mouth/feet/udder

☐ Anorexia   ☐ Neurological signs

☐ Respiratory signs   ☐ Diarrhoea

☐ Other (specify) _____

4.2 Autopsy findings – please attach results *(if applicable)*

4.3 Sample collection *(if applicable)*

Sample ID: _____

Laboratory: _____

Sample Type: _____

Submission Date: _____/_____/_____

4.4 Laboratory findings – please attach results *(if applicable)*

Section 5: Environment

5.1 Animal husbandry in outbreak area *(choose all applicable answers)*

☐ Farm   ☐ Pen or stable

☐ Grazing in defined area   ☐ Free-grazing

☐ Other (specify) _____

5.2 Please provide details of any shared water sources within the outbreak area:

_____

5.3 Name all livestock markets, slaughterhouses and animal collecting centres within a 10 km radius of the outbreak *(if applicable)*

Section 6: Risk factors and aetiology

6.1 Have animal herds within the outbreak area received vaccination?

☐ No

☐ Yes

Date of vaccination _____/_____/_____

Lot _____

6.2 Have animal herds within a 5km radius of the outbreak area received vaccination?

☐ No

☐ Yes

Date of vaccination _____/_____/_____

Lot _____

6.3 Movement of possible reservoirs

Type of possible reservoir	Date of movement	Origin	Destination
Animals:			
Carcass or meat product:			
Animal feed:			
Farmers, traders or other people:			
Vehicles:			
Others:			

6.4 If an outbreak of this nature has occurred within a 10km radius previously, please provide details:

i)   Date of last outbreak _____/_____/_____

ii) Location of last outbreak _____

iii) Disease and serotype confirmed by laboratory *(if applicable)*_____

Please attach map(s) of outbreak location, water sources, livestock markets, slaughterhouses and animal collecting centres within a 10km radius of the outbreak area

B. Foot-and-mouth disease

B.1 Aetiology

The FMD virus is a member of the Aphthovirus genus of the family Picornaviridae. The virion is non- enveloped, about 25 nm in diameter, and has an icosahedral symmetry. It contains a molecule of single-stranded RNA and 60 copies of each of the four structural polypeptides (VP1, VP2, VP3 and VP4). Of these, VP1 contains antigenic determinants that are important in stimulating neutralising antibodies in infected hosts.

There are seven serotypes of FMD virus: A, O, C, SAT 1, SAT 2, SAT 3 and Asia 1. All the serotypes produce a disease that is clinically indistinguishable but immunologically distinct. There is no cross- immunity among serotypes. Within each serotype there is a spectrum of antigenic variation, with some strains being closely related to each other and others only distantly. Antigenic variation tends to be greatest within type A. Analysis of strains of FMD virus by antigenic and genetic profiles is important in epidemiological studies and for the selection of the most appropriate vaccine strains for a region where vaccination is practised.

At temperatures below freezing, the virus is stable almost indefinitely. Even at 4 °C in simple media the virus retains infectivity for more than a year. Suspensions of virus will retain infectivity for eight to ten weeks at ambient temperatures of approximately 22°C, and for up to ten days at 37 °C. Above this temperature, inactivation is more rapid.

Sunlight per se has little effect on the virus. Environmental inactivation is related more to the effects of desiccation (less than 60 percent relative humidity) and temperature. Acid and alkaline formulations are the most effective methods for disinfection.

B.2 Epidemiology

Of the domestic livestock species, cattle, water buffaloes, pigs, sheep, goats and deer are susceptible to FMD; the disease is generally most severe in cattle and pigs. Camelidae (camels, llamas and vicuñas) have a low susceptibility. Wild cloven-hoofed species are susceptible. Though rare, FMD in elephants, hedgehogs and some rodents has been documented. Some FMD virus strains have a more pronounced predilection for one livestock species or another (e.g. pigs or cattle).

Worldwide distribution

FMD is endemic and at a high prevalence in many countries in Africa, the Middle East and Asia and is also present in parts of South America. Europe, North and Central America, the Pacific nations and the Caribbean are free of the disease. Table 4 shows the worldwide distribution of the various serotypes (since the early 1990s).

Table 4: Worldwide distribution of foot-and-mouth disease serotypes.

Serotype	Distribution
Type O	Asia, parts of Africa and South America, with recent incursions into the United Kingdom and western Europe
Type A	Asia, parts of South America and Africa
Type C	South Asia and eastern Africa
Type SAT 1	Africa and the Arabian Peninsula
Type SAT 2	Africa and the Arabian Peninsula
Type SAT 3	Southern Africa
Type Asia 1	Asia and south-eastern Europe

Virus survival

The FMD virus can retain infectivity for considerable periods in the environment, provided it is protected from desiccation, heat and adverse pH conditions. For example, the virus may survive for 14 days in dry faecal material; six months in slurry in winter; 39 days in urine; 28 days on the surface of soil in autumn; and three days on the surface of soil in summer. Such observations have generally been made in countries with a temperate climate, and these times can be expected to be much shorter in countries with hot climates.

The respiratory system is the major route of infection in ruminant species, and very small doses of virus can initiate infection. The respiratory route is also the usual portal of entry for pigs, but these animals are much more susceptible to infection by the oral route than ruminants. The virus can also enter through abrasions in the skin or the mucosae as a result of injury caused by damage from grass seeds, feeding on rough fodder, foot rot, trauma from milking machines or from fingernails during nose restraint of cattle.

The virus is excreted in large quantities in expired air, in all secretions and excretions (including milk and semen) and from ruptured vesicles. Pigs liberate vast quantities of virus in their expired breath - about 3000 times as much as cattle.

Excretion of the FMD virus can begin up to four days before clinical disease becomes apparent, and this is of great epidemiological significance. Most excretion of the virus ceases four to six days after the appearance of vesicles, when circulating antibodies develop. The virus tends to persist in foot lesions for a day or two longer than in mouth lesions, so foot lesions may be a better source of virus for diagnostic purposes in older cases. The FMD virus has been detected in the milk and semen of experimentally infected cattle for 23 and 56 days, respectively. After recovery, up to 80 percent of ruminant animals may become persistently infected. This situation is termed the 'carrier state' and is defined as carriage of the virus beyond 28 days after infection.

The maximum reported carrier periods for different species are: three and a half years for cattle; nine months for sheep; four months for goats; and five years or more for African buffaloes. The virus can be recovered intermittently from such animals by oesophageal-pharyngeal probang collections. The quantity and frequency of virus that can be collected decline progressively with time. Pigs do not become long-term carriers and cease excreting the virus within three to four weeks of becoming infected.

Disease transmission

Pigs are regarded as important amplifying hosts for the disease because of their ability to be infected orally and their capacity to excrete large quantities of virus in their exhaled breath. Cattle are regarded as good indicator hosts because of their extreme sensitivity to infection by the respiratory route, and the usual development of severe, classical clinical signs. Sheep have been thought of as maintenance hosts, because infection with some virus strains can spread through flocks with little overt sign of disease. It must be stressed that not all FMD viruses will behave in the same way epidemiologically nor will they all have the same host range. Transmission occurs by: (1) direct contact, (2) indirect transmission, (3) swill feeding of pigs, (4) windborne spread, and (5) artificial breeding.

The epidemiological pattern of the disease tends to be different in temperate and tropical or semitropical parts of the world. In the former, the greater survival of the virus in the environment means that indirect transmission through fomites may be as important as direct contact between infected and susceptible animals. Windborne virus spread is possible under some environmental circumstances. On the other hand, in hotter climates, indirect means of transmission assume less relative importance than direct means of transmission. It is often the movement of potentially infected animals and livestock trading patterns that provide the key to understanding the epidemiology of FMD in such areas.

Direct transmission of FMD arises from contact with the following material from infected animals:

– Ruptured vesicles

– Faeces

– Urine

– Breath

– Saliva

– Milk

– Windborne spread of virus from infected farms

Indirect transmission of FMD arises from contact with the following material that has been in contact with infected animals:

– Meat, blood, offal, hides

– Vehicles

– Animal bedding

– Clothing, footwear

– People (particularly farmers and veterinarians)

– Animal feed (e.g. hay)

Incubation period

The incubation period in cases of naturally acquired disease is variable and depends mainly on the animal species, strain of virus, exposure dose and route of entry. It is usually 2 – 8 days but can be longer (up to 11 – 13 days), particularly when the animal is exposed to lower concentrations of virus. Species-specific incubation periods are as follows:

– Domestic cattle: 1 – 7 days, but usually 3 – 6 days (Sard, 1978; Radostits et al., 2007). Lesions appear at 5 – 8 days after exposure to infected steers (Burrows, 1968). Incubation periods as long as 13 – 14 days have been reported (Sellers et al., 1971).

– Domestic sheep: 3 – 8 days, possibly up to 21 days. As short as 18 hours after experimental infection (Geering, 1967).

– Domestic pig: lesions appeared 7 – 13 days after exposure to infected steers (Burrows, 1968).

The incubation period for index cases in an outbreak tends to be longer than for subsequent cases;

it may be as short as 18 – 24 hours when the disease is experimentally produced.

Sub-clinical infectious period is defined as the number of days between the beginning of virus shedding and the onset of clinical signs. This is estimated to be between 2 and 5 days (Burrows, 1968).

Morbidity and mortality

The percentage of animals that become affected in a population is often high and may approach

100%. Morbidity may be lower under the following conditions:

– Where FMD virus strains are less virulent (e.g. with occult FMD strains, morbidity can be very low in some populations).

– Where species are less susceptible to disease.

– Where there is immunity, either through naturally circulating virus (in endemic regions) or through vaccination.

In domestic cattle, mortality seldom averages more than 3% and is often less than 1%. Occasionally, mortality rates may be higher, especially in juveniles. A malignant form of the disease has been reported, resulting in mortality risk approaching 50% in adult cattle (Timoney et al., 1988, Radostits et al., 2007). Complications from secondary bacterial infections may require euthanasia (Timoney et al., 1988).

In domestic pigs, mortality from FMD is usually less than 5%. It may reach 50% in piglets (Taylor, 1983). Mortality rates are higher in young pigs than in calves (Timoney et al., 1988). Mortality in unweaned piglets due to myocarditis can be up to 100% and can precede any other signs of the disease (for example, vesicles on the teats of lactating sows).

Usually, the mortality rate in adult sheep (Martin and Aitken, 2000) is zero. Mortality in lambs may be very high (Martin and Aitken, 2000).

Immunity

Circulating neutralising antibodies develop within four to ten days of infection. Convalescent animals usually have a very long immunity to reinfection (as long as five years) with closely related virus of the same serotype, but remain fully susceptible to other serotypes.

The degree of protection after vaccination is greatly influenced by the antigenic relationship between the vaccine strain and the challenge strain. Vaccines provide only partial immunity against antigenic variants of the same serotype. Potent vaccines confer immunity as early as four days after injection. However, vaccine immunity is not long lasting and therefore revaccination at regular intervals (e.g. every 6 – 12 months) is required.

Manufacturers of commercial FMD vaccines normally recommend a primary immunisation regime of an initial dose followed within three to four weeks by a second dose of vaccine. However, in endemic situations it is more usual to give two doses at six months apart and to revaccinate thereafter at yearly intervals. A proportion of vaccinated animals, although protected against the clinical disease, may become sub-clinically infected after natural challenge and excrete virus. It is important to note that animals incubating the disease when vaccinated may still develop the disease, sometimes in a milder form, and that vaccinated, exposed animals may still transmit infection for 7 – 14 days after vaccination and exposure.

B.3 Diagnosis

Antigen detection

The two most commonly used tests are: (1) the complement fixation test (CFT), and (2) the ELISA (an indirect sandwich test). The ELISA test has largely replaced the CFT. The latter is sensitive and easier to apply but, as with all tests, needs to be properly standardised to optimise its sensitivity and specificity.

Antibody detection

Serological tests for FMD include:

- The virus neutralisation test (VNT): this is a sensitive serotype-specific test, which requires three days to provide a result.

- The ELISA test (liquid-phase or solid-phase blocking): this is another sensitive serotype-specific test. It is now widely used because it provides fast results and, unlike the VNT, does not require elaborate tissue culture laboratory facilities. Positive results can be obtained within five hours of the laboratory receiving the sample.

- ELISA tests to detect antibodies against FMD non-structural proteins (NSP): the preparation of modern FMD vaccines results in the depletion of NSP. Sera from vaccinated animals contain antibodies against structural proteins, but not against NSP such as 3ABC or 2C. ELISA tests for NSP antibodies are major FMD diagnostic advances as they allow antibody titres that result from FMD infection to be distinguished from those resulting from vaccination.

Both the VNT and ELISA are OIE-prescribed tests for international trade. Reverse transcription (RT) polymerase chain reaction (PCR) tests are available for FMD. PCR is a highly sensitive and specific technique but, because of the possibility of cross-contamination, as well as expense, its use is almost entirely confined to specialist laboratories.

B.4 Control and eradication

The aim of control measures is to break the transmission between infected and susceptible animals. To be effective, all routes of transmission must be considered and addressed with controls and biosecurity measures, some examples include:

– Stopping animal movements into and out of an infected village.

– Disinfecting all vehicles before leaving a restricted (infected) area.

– Not allowing animal feed produced in an affected area to be fed to susceptible animals in an unaffected area.

– Ensuring that people follow strict biosecurity and disinfection procedures before leaving an infected area.

– Using vaccination to reduce the number of susceptible animals in an infected area.

B.5 Clinical signs

Cattle

FMD has an incubation period of 2 to 14 days, depending on the infecting dose, strain of virus and susceptibility of the individual host. The following clinical signs may be seen:

> **Specific clinical signs.** Vesicles on the tongue, hard palate, dental pad, lips, gums, and muzzle. Vesicles on the coronary band and interdigital space. Vesicles on the teats, particularly in lactating cows. Profuse salivation. Nasal discharge which is mucoid at first then becoming mucopurulent. A dramatic drop in milk yield.
>
> **Less-specific clinical signs.** Pyrexia lasting 1 to 2 days (rectal temperatures of 40 °C). Mortality in young calves. Inappetence and weight loss. Stamping of feet and lameness. Stock prefer to lie and resist attempts to raise them. Lactating cows with teat lesions, mastitis and difficulty in milking.
>
> FMD should be suspected if multiple animals in a herd have the clinical signs listed above.

| (a) Foot lesions | (b) Tongue lesions |

Figure 23. Clinical signs of foot-and-mouth disease in cattle.

Pigs

The incubation period of FMD in pigs varies with the strain of infecting virus, the dose of virus, the route of infection, individual susceptibility and the environment under which the animals are kept. It is usually two or more days, but can be as little as 24 hours (particularly when infection is spreading within a herd).

Specific clinical signs. Initially mild lameness and blanching of skin around the coronary bands. Vesicles on the coronary band and heel of the feet (including the accessory digits), on snout, lower jaw and tongue. Lesions on the coronary band are the most consistent sign of FMD in pigs. Vesicles on the tongue are usually found far back on the tongue or very small vesicles of erosions are found at the tip. If the pigs are housed on hard floor surfaces, there may be evidence of vesicles on the elbows, hocks or other areas of prior skin abrasion. Teat lesions in lactating sows.

Less-specific clinical signs. Pyrexia (rectal temperatures 39 to 40 ˚C). Lethargy, groups of pigs huddle together. Reduced appetite, inappetence. Mortality in young piglets. Stock prefer to lie and resist attempts to raise them.

FMD should be suspected if multiple animals in a herd have the clinical signs listed above.

(a) Snout lesions

(b) Foot lesions

Figure 24. Clinical signs of foot-and-mouth disease in pigs.

Small ruminants

The incubation for FMD virus in sheep is usually between 3 and 8 days. Vesicular disease may fail to develop in some infected sheep, while others may show only a single visible lesion. A notable feature of FMD in small ruminants is that the clinical signs of disease are subtle and often unapparent. Thorough clinical examination of 10 to 20 animals in a flock is often required to make a diagnosis.

Specific clinical signs. Vesicles develop on the interdigital cleft, bulbs of the heel and coronary band. These lesions can be very difficult to see. Vesicles form in the mouth, but they rupture more easily and are usually only seen as shallow erosions, most commonly on the dental pad, but also on the hard palate, lips and gums. Vesicles can appear on the teats or prepuce.

Less-specific clinical signs. Lameness is usually the first sign of FMD in sheep and goats. In a field situation, lameness due to other causes may already be present and this may conceal the presence of FMD. Reluctance to walk. Mortality in lambs and kids.

FMD should be suspected if multiple animals in a herd have the clinical signs listed above.

(a) Ruptured vesicle on dental pad

(b) Lesion on interdigital space and coronary band. Note the blanching and swelling

Figure 25. Clinical signs of foot-and-mouth disease in sheep.

B.6 Ageing lesions

When investigating an outbreak of FMD, it is possible to make relatively accurate estimates of the age of lesions during a clinical examination of affected stock. This is important, because it allows you to estimate the date of onset of clinical signs in the herd (or flock). This, in turn, then allows you to estimate the likely date of exposure to virus.

Information can usually be obtained from the herd/flock manager to indicate when clinical signs first appeared. When there is some uncertainty in this estimate, or in situations where the animals have not been closely monitored, ageing of the lesions can be a valuable tool. For example, if you are visiting a village in which there are four farms and all of them are infected with FMD, you can examine animals on all of the farms and estimate the age of their lesions, and the farm that has animals with the oldest lesions can be called the index case farm.

Ageing lesions is particularly useful for a disease such as FMD because the clinical progression of lesions in affected animals follows a relatively predictable course. The table and the graph below can be used as a reference. It is important to bear in mind that the ageing of lesions is approximate because other factors (such as the presence of secondary infection) can alter the rate at which the lesions heal.

Table 5: Description of the clinical appearance of FMD lesions according to the number of days post infection.

Day	Appearance
1	Blanching of epithelium followed by formation of fluid filled vesicles.
2	Freshly ruptured vesicles characterised by raw epithelium, a clear edge to the lesion and no deposition of fibrin.
3	Lesions start to lose their sharp demarcation and bright red colour. Deposition of fibrin starts to occur.
4	Considerable fibrin deposition has occurred and re-growth of epithelium is evident at the periphery of the lesion.
7	Extensive scar tissue formation and healing has occurred. Some fibrin deposition is usually still
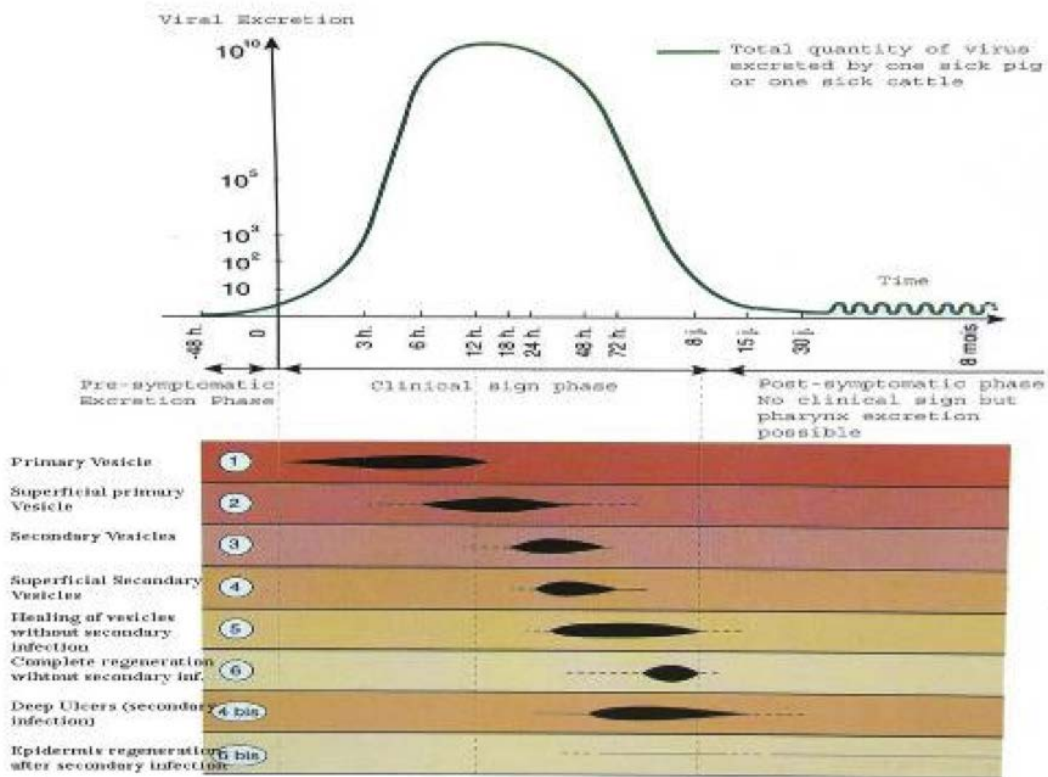
Figure 26: Diagram showing the pattern of appearance of FMD lesions and their relationship with viral excretion as a function of time.

Note: Detailed information on FMD, including information on aetiology, epidemiology, diagnosis, prevention and control, can be found in:

OIE (2017). Terrestrial Animal Health Code. World Organisation for Animal Health. Available at: http://www.oie.int/en/international-standard-setting/terrestrial-code/access-online/

OIE (2017). Manual of Diagnostic Tests and Vaccines for Terrestrial Animals. Available at: http://www.oie.int/en/international-standard-setting/terrestrial-manual/access-online/

OIE Technical disease cards: http://www.oie.int/animal-health-in-the-world/technical-disease-cards/

C. Classical swine fever

C.1 Aetiology

Classical swine fever (CSF) is caused by a *Pestivirus*. Although there is only one antigenic type, there are a number of different strains which vary in their virulence. The virus is capable of surviving for long periods of time in frozen, salted or smoked pork. This is an important characteristic that increases its potential for geographical spread, as virus can easily be transferred from one place to another in pork products and cause disease outbreaks in pigs fed uncooked garbage.

C.2 Epidemiology

Worldwide distribution

The disease is enzootic in Asia, continental Europe, South America and Central America. Canada, the USA and the Caribbean, apart from Cuba, are free from the disease. Apart from Madagascar and Mauritius, Africa is also free from the disease. In Oceania, the disease has only occurred on two occasions (New Zealand in 1930 and 1953), but on both occasions the disease was quickly eliminated by slaughtering the affected herds.

Virus survival

CSF virus is moderately fragile in the environment. This virus is sensitive to drying and ultraviolet light and is rapidly inactivated by a pH less than 3. Sodium hypochlorite and phenolic compounds are effective disinfectants. CSF virus can survive for long periods in meat, but is destroyed by cooking.

Disease transmission

Infection of susceptible pigs is normally by the oral or nasal routes. Even before clinical signs develop, infected pigs shed virus in all their excretions and secretions. The normal means of spread is by direct contact between infected and susceptible animals, but airborne infection can occur over short distances. With the chronic form of the disease, virus persistence is characteristic. Infection of pregnant sows with low-virulence strains often results in congenital infection in their piglets. Such infected piglets are immunotolerant virus shedders and are often responsible for maintaining the disease in a herd. Feeding infected pork products to pigs (i.e. garbage feeding) has been an important way of keeping the disease cycling in enzootic areas or introducing disease into previously free areas. Humans can spread infection by syringes, vaccination paraphernalia and the like. Flies may transmit infection mechanically to nearby piggeries.

As stated above, CSF can enter a country by means of infected pork products finding their way, un- treated, to pigs fed garbage. This is what happened in the two outbreaks that have occurred in New Zealand. Many countries have implemented legislation to ensure that garbage fed to pigs must first be cooked so that the risk of introduction by means of meat infected with CSF virus (or other exotic infectious agents) is lessened. Legally imported pork may only come from countries that are free from the important exotic diseases of pigs, so the risk is low, but smuggled pork products represent a much greater risk of introducing CSF (and FMD and swine vesicular disease) viruses. Quantitatively, the amounts of smuggled pork products entering the country are small, most are eaten by humans, and any waste is more likely to be disposed of in urban dumps than in garbage collected for feeding to pigs. Nevertheless, some risk probably exists and, for this reason, piggeries which feed garbage to their animals continue to be regulated and kept under surveillance.

It is likely that any introduction of CSF would first be evident in garbage-fed pigs. The classical acute disease would quickly be brought to the attention of a veterinarian, allowing rapid action to be taken. The danger with less-virulent disease is that it may be allowed to carry on for some time before a diagnosis is sought, and during that time the opportunity for spread to other herds may be increased by movement of infected pigs. The worst scenario is where the disease establishes itself in a wild pig population.

Direct transmission of CSF arises from contact with the following material from infected animals:

– Nasal and/or lacrimal secretions

– Urine

– Faeces

– Uterine fluids

Vertical (trans-placental) **transmission**

Indirect transmission of CSF arises from:

– Consumption of infected meat

– Artificial insemination

– Contact with contaminated mechanical vectors, including personnel, vehicles, contaminated clothing, other animals and arthropods

Breeding herds are very important in the control of CSF. Animals in an infected breeding herd will often show only subtle signs of disease, but they are an important source of infection (through production of congenitally infected piglets, for example). Breeding herds are also important because they will often supply pigs to other farmers or producers, thus facilitating the spread of disease. Breeding herds should be a main target in a control programme.

Village smallholdings will not usually be the source of an outbreak of CSF. Smallholdings usually tend to become infected when infected pigs are bought from commercial breeders. But smallholdings can represent a risk if smallholders practise swill feeding (which increases the risk of pigs becoming infected by the oral route) or if there is the opportunity for pigs to come into contact with wild pigs.

Incubation period

The incubation period for CSF varies from 2 to 14 days. However, this is complicated by the fact that there are several different forms of the disease. When you are performing tracing during investigation of an outbreak of acute or peracute CSF, you should use the incubation period of 2 – 14 days, but be aware of the possibility of sub-clinically infected herds being a source of CSF virus.

Morbidity and mortality

There are three forms of CSF. Which form an animal or herd develops depends on the time of infection (pre-natal or post-natal) and the virulence of the viral strain:

– Acute: post-natal infection with highly virulent strains.

– Chronic: post-natal infection with low-virulence strains.

– Congenital infection: pre-natal infection with low-virulence strains.

Highly virulent strains result in a very contagious viral infection and large amounts of virus excretion. Virus is shed in oronasal and lacrimal secretions, urine and faeces. There is rapid spread, mainly through direct pig-to-pig contact. The virus can also be transmitted indirectly by mechanical vectors such as people, vehicles, other animals and arthropods. Pig density plays an important role in the transmission of highly virulent strains of CSF.

Infection with low-virulence strains of CSF will often go unnoticed, and disease is transmitted from sows to piglets by in utero transmission. There may be a short period of virus excretion during the acute phase of infection, but with lower levels of virus than the highly virulent strains. A large quantity of virus is shed during farrowing, and congenitally infected piglets represent a continuous source of infection. With the chronic form of the disease there is clinical improvement after the acute

phase of disease, but a persistent leukopenia remains. A second period of illness then follows, characterised by anorexia and depression, fever and, often, death. Pigs that survive have growth retardation, skin lesions and arched backs. They may continue to live for 100 or more days, but will eventually die. These animals will be seropositive if serological testing is carried out.

The carrier state is a very important component of the epidemiology of CSF and plays a major role in the maintenance of the virus in a population. The carrier state occurs when a piglet is infected in utero at such a time that the piglet is immunotolerant to CSF virus. The immunotolerant pig appears healthy at birth. These apparently healthy 'carrier' piglets continuously excrete virus and can live up to 11 months of age. They are not easy to detect and so remain a constant source of infection for susceptible pigs throughout this time, thus perpetuating CSF in the population. It is therefore important to ensure that control measures prevent congenital infection of pigs.

C.3 Diagnosis

When virulent virus strains are involved, the clinical picture in an infected herd will be strongly suggestive of the disease. With less-virulent strains, care must be taken that the disease is not ascribed to other causes, such as salmonellosis, even though such bacteria may be present. The disease may be difficult to differentiate from African swine fever, which presents with similar clinical signs.

Virus isolation provides the definitive diagnosis. Virus can be recovered from a number of tissues, though tonsil is the preferred tissue. Immunostaining (fluorescent antibody or immunoperoxidase) is a valuable method to demonstrate virus in tonsil tissue. However, care must be exercised to use CSF-specific monoclonal antibodies for such tests, as infection of pigs with bovine viral diarrhoea virus or border disease virus will cross-react with classical swine fever antisera.

C.4 Control and eradication

During outbreaks, confirmed cases and contact animals may be slaughtered and a quarantine imposed. Destruction of infected herds and subsequent cleaning and disinfection of premises should be straightforward. Carcasses derived from infected premises prior to the diagnosis being made, would be traced and destroyed to prevent any spread of infection by garbage feeding. Providing diagnoses are made promptly, inter-herd spread should be limited and the possibility of wild pigs becoming infected much reduced.

Vaccination may be used as a tool to assist in controlling an outbreak and eradicating the disease. In countries where the risk of reintroduction of virus is high, control by vaccination is the control method of choice. Periodic serologic sampling is necessary to monitor for the potential reintroduction of disease. Modern vaccinations allow a distinction to be made between infection and vaccination titres.

It is likely, in the initial stages of an eradication response, that a controlled area would be established within which movement of live pigs and pork products would be restricted and pig herds monitored for any spread of disease. The extent and duration of these measures would be decided upon following an assessment of the time between infection and detection of the index case and would be governed by factors such as: the presence/absence of any secondary outbreaks; the destinations of animals moved from infected herds in the period before diagnosis; and the distribution of pork product from abattoirs that had processed pigs from the infected herds prior to diagnosis. New stock could be introduced to properties 15 days after cleaning and disinfection.

C.5 Clinical signs

CSF is a highly contagious viral disease affecting pigs and wild boar. The disease can occur in a number of forms (acute, chronic and congenital). Many of the signs are relatively non-specific, so it is important to know some of the major differential diagnoses of this disease. Given the seriousness of CSF and of some of its differentials, the appearance of clinical signs that

are typical of CSF in a number of animals warrants further investigation and implementation of emergency control measures. The differential diagnoses for CSF include:

– African swine fever (cannot be clinically distinguished)

– Salmonellosis

– Erysipelas

– Pasteurellosis

– Viral encephalomyeletis

– Thrombocytopaenia purpura

– Warfarin poisoning

Acute CSF

With the peracute form of the disease, death occurs within 24 to 48 hours of infection. Mortality can reach 100%. Often, the only clinical finding is sudden death.

Clinical signs of the acute form of disease include: inappetence/anorexia; discharge from the eyes and nose; enlarged lymph nodes; coughing; constipation followed by diarrhoea; fever (usually 40 C); huddling; haemorrhages (redness) on the skin of the ears, tail, abdomen, and inner surfaces of the limbs; weight loss; staggering gait; cyanosis (blue colour) of the ears and nose; and convulsions.

Post-mortem findings with the acute form of disease include: petechial (small) haemorrhages on the epiglottis; enlarged and haemorrhagic lymph nodes; enlarged and necrotic tonsils with pinpoint haemorrhages, petechiae and ecchymoses on the skin; haemorrhages in the body organs, particularly the kidney, heart, bladder, lung, gall bladder, and spleen; oedema of the lungs; fluid in the body cavities; and encephalomyelitis with perivascular cuffing.

Sub-acute infections can occur in which the signs are the same as the acute form but generally milder, with a lower rate of mortality.

Chronic CSF

Pigs with the chronic form of CSF will often appear normal and show good appetite for a longer period of time than those with the acute or sub-acute form of the disease. Growth retardation and wasting are the most evident signs.

Clinical signs of the chronic form of disease include: fever; failure to thrive; diarrhoea; difficulty breathing; coughing; dermatitis (which is often generalised); reddening of the skin; and abortions.

Post-mortem findings with the chronic form of disease include: enlarged lymph nodes; atrophy of the thymus; inflammation around the heart and the lining of the chest cavity; consolidation of the lungs; poor body condition; small (button) ulcers in the large intestine; secondary pneumonia; and enteritis.

Mild or sub-clinical CSF

This form of the disease is typically seen in sows that have been inadequately vaccinated, or sows which become infected with a virus of low virulence. These sows may appear normal, but give birth to shaking piglets, many of which die.

Clinical signs of the mild or sub-clinical form of disease include: transient pyrexia; transient inappetence; foetal death; foetal resorption; mummification; stillbirth; or birth of live, congenitally deformed piglets. Abortion rarely occurs.

Detection of CSF in pig-breeding operations can be particularly difficult, since clinical signs in adult pigs may be mild and similar to other (more common) diseases. CSF should be suspected in any case of reduced fertility in a herd, especially where

other risk factors (e.g. the presence of disease in wild boar) are present and/or other diseases of the reproductive tract have been excluded.

Congenital CSF

Clinical signs of the congenital form of disease include: congenital tremor; weakness; runting; and poor growth.

The congenital form of CSF occurs when a sow is infected during pregnancy. The sow may or may not show any sign of disease. If the sow is infected before the piglets' immune system is developed (from 50 to 70 days of gestation), the piglets will be apparently healthy at birth. These piglets shed virus after they are born and therefore represent a risk to susceptible pigs or susceptible herds. At several weeks of age these piglets develop typical clinical signs of CSF, but these are likely to be milder than usual, last longer and there is no associated fever. If a pregnant sow is infected before 50 days of gestation, foetal death, mummification, abortion and deformity of piglets typically results.

Note: Detailed information on CSF, including information on aetiology, epidemiology, diagnosis, prevention and control, can be found in:

OIE (2017). Terrestrial Animal Health Code. World Organisation for Animal Health. Available at: http://www.oie.int/en/international-standard-setting/terrestrial-code/access-online/

OIE (2017). Manual of Diagnostic Tests and Vaccines for Terrestrial Animals. Available at: http://www.oie.int/en/international-standard-setting/terrestrial-manual/access-online/

OIE Technical disease cards: http://www.oie.int/animal-health-in-the-world/technical-disease-cards/

D. Equipment checklist

The following equipment should be ready for use at all times and should be taken by the investigation team or individual investigator when carrying out a field visit.

D.1 Personal protective equipment

– Latex or plastic gloves

– Protective mask

– Protective eye glasses

– Boots

– Protective suit

D.2 Restraint equipment

– Ropes

– Pig restrainers

D.3 Disinfectant and cleaning material

– Brush

– Soap

– Detergent

– Cotton wool

– 70% alcohol solution

– Disinfectant solution, e.g. 4% washing soda (Na_2CO_2), soap, 1 – 2% sodium hydroxide solution

– Spraying equipment, footbath equipment

D.4 Medical supplies

– Antibiotics and antiseptics, e.g. penicillin, gentian violet, 70% alcohol solution, Negasunt dusting powder

– Vaccine (if available) with appropriate cold chain facilities

– Cotton, gauze, and tape

– Forceps

– Sedative or anaesthetic drugs

D.5 Sample collection

– Tube with glycerine buffer

– Needles and syringes

– Plastic bags and rubber bands

– Cooler with ice packs

– Cotton swabs

– Sticky labels

D.6 Data collection

– Outbreak investigation forms

– Case recording forms

– Sample submission forms

– Notepad and protective folder

– Pens and pencils

– Recording tape

– Other necessary stationery, e.g. tape and glue, scissors, stapler, batteries

D.7 Communication, transportation, navigation

– Mobile phone or radio communication equipment

– List of all field stakeholders

– Public awareness material

– Digital camera

– GPS device

– Detailed maps of affected area

– Gasoline

This list should be photocopied so that it is available for use by all who work in your office.

E. Cleaning and disinfection

Decontamination[1] is the combination of physical and chemical processes that kill or remove pathogenic microorganisms. Decontamination of premises where infected animals have been housed reduces the risk of the spread of infectious disease to new animals. When premises have been identified as infected and measures have been undertaken to remove or eliminate the source of infection on those premises, disinfection needs to be carried out. The disinfection process should include items such as:

– Buildings with wooden, metallic or masonry structures

– Machinery of mostly metallic components

– Pipework of various types

– Water tanks

– Animal food storage areas

– Sewage waste

Depending on the infectious agent involved, different decontamination procedures and disinfectants need to be used for different sites on the premises. In situations where the disease agent does not spread directly from animal to animal (e.g. bluetongue), comprehensive decontamination of the premises is not warranted. In contrast, some viruses (e.g. swine vesicular disease and foot-and-mouth disease) are relatively stable on inanimate objects and can be spread to remote animals via contaminated people, clothes and equipment. Viruses that can be spread by this type of contact require the most comprehensive decontamination programmes.

Preliminary cleaning work is invariably required before any chemical disinfectants are applied. Simple cleaning of surfaces by brushing with a detergent solution is effective in removing contaminating viruses and is fundamental for achieving effective chemical decontamination. Most disinfectants have reduced effectiveness in the presence of fat, grease and organic dirt. Every effort should be made to remove organic matter from all surfaces that are to be decontaminated. Hot water and steam are effective in cleaning many cracks and crevices where pathogens are likely to accumulate. The insides of pipework can often only be cleaned effectively by steam.

A knowledge of the properties of the contaminating virus is a fundamental part of planning a decontamination strategy. Choosing the most appropriate disinfectant is dependent on the nature of the virus particles. Three general classes of virus have been described (Klein and DeForest, 1981).

–   Category A viruses are of intermediate to large size and contain lipid, which makes them susceptible to detergents, soaps and most disinfectants. Such viruses are susceptible to dehydration and do not persist, except in cool, moist environments. The best disinfectants to use for these viruses are detergents, hypochlorites, alkalis, Virkon or glutaraldehyde (see below).

–   Category B viruses have no lipid and are smaller and more hydrophilic. These viruses are relatively resistant to lipophilic disinfectants such as detergents. Although they are sensitive to most disinfectants they are less susceptible than viruses in Category A. The best disinfectants for these viruses are hypochlorites, alkalis, Virkon or glutaraldehyde (see below).

–   Category C viruses have no lipid and are intermediate in size. These viruses fall between Categories A and B in terms of their sensitivity to disinfectants. The best disinfectants to use for these viruses are detergents, hypochlorites, alkalis, Virkon or glutaraldehyde (see below).

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1 This section of the manual is a summary of information presented in Geering et al. (2001) and in the Handbook of the Philippines National Foot-and-Mouth Disease Task Force 1995 – 2003

E.1 Types of disinfectants

Disinfectants can be grouped into five chemical categories:

– Soaps and detergents

– Oxidising agents

– Alkalis

– Acids

– Aldehydes

Soaps and detergents

The application of soaps and detergents is the essential first step for achieving decontamination. In most cases, the primary aim is the removal of organic material, dirt or grease from the surfaces that are to be decontaminated. Most industrial and domestic brands of soaps and detergents are satisfactory. Hot water, brushing and scrubbing enhance the cleaning action. Similarly, steam improves the cleaning and decontamination process by raising the temperature and penetrating crevices. However, steam by itself can only be used as a decontaminant if the temperature of the surface can be raised to at least 100 C and held there long enough to allow inactivation of the agent. Because of uncertainties regarding temperatures and times of contact, steam is only recommended as an adjunct to decontamination.

The surfactant action of soaps and detergents is an effective decontaminant for all Category A viruses, because of their outer lipid envelope. For decontamination procedures involving exotic viruses in Category A, soaps and detergents are effective disinfectants in their own right.

Many commonly used disinfectants in hospitals, surgeries, dairies and food-processing areas involve soapy combinations of phenolics or quaternary ammonium compounds. These agents are specifically antibacterial and are effective against category A viruses. They have no activity against category B viruses and limited activity against category C viruses. Although they may be useful for preparatory cleaning purposes during an outbreak of an exotic viral disease, they are not recommended because more effective cleaning agents and viral decontaminants are available.

Iodophors are combinations of solubilising agents and a carrier that releases free iodine. It is difficult to define active concentrations with certainty in all circumstances, so iodophors are not recommended for the inactivation of viruses.

Oxidising agents

Oxidising agents are the disinfectants recommended for most situations. Chlorine is released from hypochlorite solutions and is a powerful oxidising agent that is effective against all virus groups. The effectiveness of hypochlorite is greatest in the pH range 6 -– 9. The activity of hypochlorite decreases markedly in the presence of organic material. Hypochlorite solutions are not chemically stable and decompose rapidly as temperatures rise above 15 C.

Virkon is a disinfectant with outstanding virucidal properties. It has low toxicity and is effective against members of all 17 virus families, but it has not been approved for use on skin. Its activity is based on a buffered synergised acid peroxygen system containing a high percentage of surfactant. It is relatively safe to use and comes in a powdered form ideal for dilution at the site of an exotic disease outbreak. It can be sprinkled in powdered form over wet or boggy areas, but the concentration of disinfectant achieved in this situation cannot be accurately controlled.

Alkalis

Both sodium hydroxide (caustic soda) and sodium carbonate (washing soda) are widely available in large quantities at low cost and both have a natural saponifying action on fats and other types of organic matter, which assists the cleaning process. Because they are virucidal under heavy burdens of organic material, they are ideal agents for decontaminating animal housing, yards, drains, effluent waste pits and sewage collection areas.

Acids

Acids are generally highly virucidal. They have a wide range of uses, ranging from the treatment of liquid effluent to personal decontamination. Hydrochloric acid is a strong acid that is widely available from hardware stores and is less toxic than other strong acids. Citric acid is a milder acid, available in solid form, that is active against acid-sensitive viruses and can be used safely for personnel and clothing decontamination. It is particularly useful when added to detergents for the inactivation of foot-and-mouth disease virus.

Aldehydes

Glutaraldehyde is an effective disinfectant that is active against all virus families and other microorganisms in concentrations of 0.2% w/v. It remains effective in moderate concentrations of organic material, is chemically stable and only mildly corrosive for metals. For large-scale decontamination, however, the cost of glutaraldehyde is likely to be high.

A 40% aqueous solution of formaldehyde gas is called formalin and is a useful disinfectant. Formalin diluted with 12 parts water produces 8% w/v formalin which is an active disinfectant against most virus families.

Gaseous formaldehyde can be used to decontaminate air spaces, the insides of motor vehicles, and equipment that must be kept dry, e.g. electronic devices. The conditions must be carefully controlled, however, in terms of gas concentration, temperature, humidity, time of contact and even distribution of the gas. Under emergency conditions on a contaminated property, it is unlikely that all of these conditions can be adequately controlled. In addition, the space to be decontaminated must be completely sealed to prevent gas escape, because the most effective dwell time for inactivation is anywhere between 15 and 24 hours. Other problems include the toxicity of gas, the dangerous nature of its generation in non-laboratory conditions (potassium permanganate reacts violently with formalin), the environmental protection guidelines that prevent the release of formaldehyde gas to the atmosphere, and the difficulty of completely purging residual formaldehyde gas from confined spaces.

E.2 Disinfectants for specific diseases

In the tables that follow, the numbers cited in the columns labelled 'Disinfectant' refer to the disinfectants listed in Table 10.

African swine fever and classical swine fever (Category A viruses)

African swine fever and classical swine fever are caused by Category A viruses (Asfaviridae and Flaviviridae, respectively). The best disinfectants to use are detergents, hypochlorites, alkalis, Virkon or glutaraldehyde.

Table 6: Recommended disinfectants for African swine fever and classical swine fever.

Item to be disinfected	Disinfectant/chemical/procedure
Live animals	Euthanasia (ASF) or vaccination (CSF).
Carcasses	Bury or burn.
Animal housing/equipment	1, 2a, 2b 2c, 3.
Environment housing/equipment	Eradicate ticks or burn wooden structures (ASF).
Humans	1.
Electrical equipment	5c.
Water tanks	Drain.
Feed	Bury or burn.
Effluent, manure	Bury or burn, 3a, 3b, 4a, 4b.
Human housing	1, 2a, 2b, 2c.
Machinery	1, 3a, 3b.
Vehicles	1, 3a, 3b.
Clothing	1, 2a, 2b, 2c, 3a, 3b.
Aircraft	1, 2c.

Avian influenza and Newcastle disease (Category A viruses)

Avian influenza is caused by a Category A virus. The best disinfectants to use are detergents, hypochlorites, alkalis, Virkon or glutaraldehyde.

Table 7: Recommended disinfectants for highly pathogenic avian influenza and Newcastle disease.

Item to be disinfected	Disinfectant/chemical/procedure
Live birds	Euthanasia or vaccination.
Carcasses	Bury or burn.
Animal housing/equipment	1, 2a, 2b, 2c, 3a, 3b.
Environment housing/equipment	N/A.
Humans	1.
Electrical equipment	5c.
Water tanks	Drain to pasture where possible.
Feed	Bury or burn.
Effluent, manure	Bury or burn, 3a, 3b, 4a, 4b.
Human housing	1, 2a, 2b, 2c.
Machinery	1, 3a, 3b.
Vehicles	1, 3a, 3b.
Clothing	1, 2a, 2b, 2c, 3a, 3b.
Aircraft	1, 2c.

Foot-and-mouth disease (Category B viruses)

Foot-and-mouth disease is caused by a Category B virus (Picornaviridae). The best disinfectants to use are hypochlorites, alkalis, Virkon, glutaraldehyde, hydrochloric or citric acid.

Table 8: Recommended disinfectants for foot-and-mouth disease.

Item to be disinfected	Disinfectant/chemical/procedure
Live animals	Euthanasia or vaccination.
Carcasses	Bury or burn, 3a, 3b, 4a, 4b.
Animal housing/equipment	2a, 2b, 2c, 3a, 3b.
Environment housing/equipment	3a, 3b.
Humans	1, 4b.
Electrical equipment	5c.
Water tanks	3a, 3b.
Feed	Bury or 5b.
Effluent, manure	Bury or 4a, 4b.
Human housing	2, 4b.
Machinery	2c, 3a, 3b, 4a, 4b.
Vehicles	2c, 3a, 3b, 4a, 4b.
Clothing	2a, 2b, 2c, 3a, 3b, 4b.
Aircraft	2c.

Rabies (Category A viruses)

Table 9: Recommended disinfectants for rabies.

Item to be disinfected	Disinfectant/chemical/procedure
Live animals	Destroy without damaging head; beware of being bitten
Carcasses	Submit head to diagnostic laboratory; burn or bury remainder of carcass
Animal housing/equipment	1 (to clean) followed by 2a, 2b
Environment housing/equipment	N/A
Humans	Bites should be thoroughly washed with 1 then cleaned with a disinfectant suitable for human wounds. Unless the animal can be shown conclusively to be free from infection, a post-exposure course of human diploid cell vaccine and human immunoglobulin should be started
Electrical equipment	N/A
Water tanks	N/A
Feed	N/A
Effluent, manure	Burn or bury
Human housing	N/A
Machinery	N/A
Vehicles	N/A
Clothing	N/A
Aircraft	N/A

E.3 Recommended concentrations and contact times

Table 10: Recommended disinfectants and concentrations for inactivation of viruses.

Disinfectant	Details	Final concentration	Contact time
1. Soaps and detergents	-	-	-
2a. Sodium hypochlorite (bleach)	1 part 5.25% sodium hypochlorite to 5 parts water.	3% w/v (30,000 ppm)	10 - 30 min
2b. Calcium hypochlorite	30 g calcium hypochlorite to 1 litre water.	3% w/v	-
2c. Virkon	20 g Virkon to 1 litre water.	2% w/v	10 min
3a. Caustic soda	20 g caustic soda to 1 litre water.	2% w/v	10 min
3b. Sodium carbonate anhydrous	40 g sodium carbonate anhydrous to 1 litre water.	4% w/v	10 min
3c. Washing soda	100 g washing soda to 1 litre water.	10% w/v	30 min
4a. Hydrochloric acid	1 part 10 Molar hydrochloric acid to 50 parts water.	2% w/v	10 min
4b. Citric acid	2 g citric acid to 1 litre water.	0.2% w/v	10 min
5a. Glutaraldehyde	As appropriate to yield 0.2% w/v.	0.2% w/v	10 - 30 min
5b. Formalin	1 part 408% w/v	10 - 30 min	
5c. Formalin gas	Only used by experienced personnel.	-	15 - 24 hours

Sodium hypochlorite: $NaClO$
Calcium hypochlorite: $Ca(ClO)_2$
Caustic soda: $NaOH$
Sodium carbonate anhydrous: Na_2CO_3
Washing soda: $Na_2CO_3 .10H_2O$
Hydrochloric acid: HCl

0.2% w/v: 2 grams (weight) per 1,000 mL (volume) water.
One part per million (ppm) is equivalent to one part per 1,000,000 parts. That is, $(1 \div 1,000,000) \times 100 = 0.0001\%$ (or 1% = 10,000 ppm).

I have a 40% w/v solution of disinfectant concentrate. The final concentration required for disinfection is 8% w/v. How many litres of concentrate are required to make 50 litres of made-up disinfectant?

A 40% w/v solution is equivalent to 400 g of disinfectant in 1 litre of water. An 8% w/v solution is equivalent to 80 g of disinfectant in 1 litre of water. How many mL of a 40% w/v solution are required to provide 80 g of disinfectant?

mL of 40% solution required = (80 × 1000) ÷ 400 mL of 40% solution required = 200

So, 200 mL of a 40% w/v solution are required to make 1 litre of an 8% w/v solution. Therefore, 10 litres (50 × 200 mL) of a 40% w/v solution are required to make up 50 litres of an 8% w/v solution.

E.4 The decontamination and disinfection process

In any large-scale decontamination and disinfection procedure the cost of soaps and/or detergents and disinfectants will be minor relative to labour and other operational costs. Use disinfectants at their recommended concentrations.

Decontamination

- Transfer animals to another place so you can clean their pens properly.

- Prepare soaps and/or detergents before you start working; wear appropriate work clothes.

- Cover all the electric outlets with plastic and masking tape.

- Remove all dirt and debris sticking to the walls and floors of pens. Remove manure, debris, bedding and other organic materials.

- Discard items that are no longer used, such as cartons, rotting wood and other materials.

- Make sure there is plenty of opportunity for water to drain away from the area in which you are working.

- Start decontamination from the top (i.e. the ceiling) then work your way downwards (to the floor).

Disinfection

- Make sure that disinfectants are freshly made up at the appropriate concentration (see Table 10 for details).

- Make sure disinfectants make physical contact with the area being disinfected for the appropriate time (again, see Table 10 for details).

- Start disinfecting from the top (i.e. the ceiling) then work your way downwards (to the floor).

- Discard unused disinfectant.

F. Sample collection and submission

Correct sample collection and submission is important in order to provide samples that generate useful information and to ensure that they are transported to the laboratory in a safe and secure manner. The following guidelines are taken from instructions provided by the Regional Reference Laboratory (RRL) for FMD in Pak Chong (Thailand). For individual country laboratory requirements, the laboratory should be contacted to supply specific instructions for collection and submission of samples.

F.1 Sample collection

Suitable samples for collection are blood, epithelium or infected tissue and vesicular fluid.

– Always use a fresh needle for every animal. Reusing the same needle can spread disease between animals and can contaminate the specimen by mixing the blood from two animals.

– Always use a new syringe for every animal. Reusing an old syringe can spread disease and contaminate the sample. Plastic syringes can be cleaned and sterilised, but they quickly become stiff and difficult to use, so reuse is best avoided.

– Restrain the animal properly before getting blood. If the animal moves, it is much harder to collect blood, and may be dangerous to the animal or to you.

– Make sure the skin of the animal is clean before taking a sample. If there is dirt or faeces on the skin, it can be scraped off and then cleaned with some alcohol on cotton wool or a swab. Let the alcohol dry before inserting the needle.

– Blood clots quickly after it has been collected. If the serological test used by the laboratory needs serum then plain blood collection tubes should be used. If plasma or blood cells are required (e.g. white blood cells for antigen detection tests) then an anticoagulant should be used to prevent clotting. Check with the laboratory which anticoagulant is preferred. When using an anticoagulant, the sample should be gently mixed once it is in the tube. Rock the tube from end to end three or four times to mix. Do not shake the tubes, as this will break the cells and ruin the sample.

– Used needles should be disposed of carefully. Never throw needles on the ground, or leave them lying around. A plastic bottle with a narrow neck makes a good container for used needles. When full, it should be burnt in a hot fire to destroy the needles.

– For jugular samples from cattle and buffalo, use a 16 to 18 gauge 1.5 inch needle. When collecting blood from the cranial vena cava in pigs, use a 20 gauge 2 inch needle (for adults). If collecting from the ear vein, a fine (19 to 23 gauge) needle should be used. For sheep and goats, collect blood from the jugular vein using a 18 to 20 gauge 1 inch needle.

F.2 Transportation and processing of samples

Blood will be in good condition for testing if three rules are followed: (a) handle blood with care, (b) do not allow blood to get too hot, and (c) do not allow blood to sit for too long before testing. When blood is collected in the field it should be placed straight into a cool box or refrigerator. If a cool box is used, sealed ice bricks should be used, otherwise the ice will melt, the tubes will get wet and the labels will run or fall off. Make sure that the tubes are kept upright at all times.

The samples can be kept in a cool box with sealed ice blocks for a single day. In a car refrigerator, they can be kept for 2 to 3 days at 4 C, but should be processed as soon as possible.

Blood is made up of blood cells and the fluid in which the blood cells are carried, the plasma. The red blood cells contain haemaglobin, which transports oxygen. If not handled properly after collection, the red blood cells can break, releasing the haemoglobin into the plasma. The red staining of the plasma makes laboratory analysis impossible. The plasma contains many different substances, including antibodies and clotting factors. When blood is collected, it can be stored in tubes either with or without an anticoagulant (such as lithium heparin, or citrate). If no anticoagulant is present the blood will clot. The

fluid that separates out from the clot is called serum. Serum is plasma without the clotting factors. The best way to make sure that blood is suitable for testing is to remove the red blood cells as soon as possible after collection. Once this is done, the serum or plasma can be frozen and stored for a very long time. Use a centrifuge to separate the cells from the serum or plasma. Place the blood tubes evenly in the centrifuge, so it is balanced, and spin them for 10 minutes at about 2000 rpm (or slightly longer for specimens with anticoagulant). When the centrifuge is finished, all the blood cells should be at the bottom of the tube, and the clear serum or plasma at the top. Use a pipette to transfer the serum or plasma from the blood tube to a serum tube. Label the serum tube, and freeze it at -20 C or colder ready for analysis.

Protocol for sample submission to the OIE RRL, Pak Chong, Thailand

It is recommended that you send samples by air freight to Suvarnabhumi International Airport, Bangkok. The Department of Livestock Development (DLD) officers will arrange the customs clearance of the samples and these will be collected by staff from the RRL. Contact staff at Pak Chong before sending samples. Packaging and dispatch of samples and biological materials should adhere to the following procedures:

1. Collection of samples.

– Epithelium and infected tissue: Vesicular fluid or epithelial tissue from vesicular lesions should be collected for laboratory diagnosis and type identification using antigen capture sandwich ELISA and virus isolation. A piece of epithelial tissue no smaller than 2 cm × 2 cm should be collected. The tissue should then be placed in a strong container, or a bottle with a screw cap, suspended in a mixture of 50% glycerin with 0.04 phosphate buffer pH 7.2 – 7.6 and added antibiotics. There will be considerable loss of infectivity if samples are sent in a buffer outside of this pH range.

– Vesicular Fluid: At least 1 mL of vesicular fluid must be collected and packed, as it is, in a tightly closed, screw-cap vial. The vial must be kept at freezing temperature if immediate transport to the FMD laboratory is not possible.

– Blood: Blood samples should be collected under sterile conditions. The serum should be separated by centrifugation soon after collection and kept in screw-cap vials with O-rings. Serum samples should be kept at -20 C before dispatch.

2. Packaging

Samples must be packed in primary and secondary IATA-approved watertight containers so that the samples arrive in good condition and do not present any hazard to persons or animals during shipment. It is essential that the contents of containers which break or leak during transit do not contaminate the outside layer of the package. The recommended procedure for packing samples is as follows:

– Samples must be put in a primary container (glass or plastic tubes or bottles) with screw caps and wrapped with paraffin film or adhesive tape individually in order to prevent leakage of fluid. The wrapping of bottles or primary containers should be carried out in clean surroundings.

– The primary container must be packed in watertight secondary packaging, which should be a strong, crushproof and leakproof metal container. The container should contain absorbent cotton wool sufficient to absorb the entire contents of the primary container.

– The secondary packaging must be placed in an outer container. This should be a polystyrene foam box covered with a hard box or IATA-approved container.

– Sufficient information and a list of samples or materials should be enclosed in an envelope, enclosed in a plastic bag and placed between the secondary packaging and outer box.

– It is recommended that the secondary packaging is placed in a freezer box to ensure that all materials are kept cool during shipment. These packs should be pre-frozen at -20 C before packaging.

– If dry ice is used for packaging, it must be placed around the outside the secondary packaging. Dry ice must not be placed within the primary packaging as it may cause breakage of the sample tubes.

3. Labelling. The outer surface of the package must be clearly labelled with the following details:

– The name and address of the institute submitting the samples.

– Contact telephone numbers.

– Infectious substance hazard label.

– light number and estimated arrival time.

– Air waybill number.

– Dry ice label (if necessary).

– The package should be addressed as follows:

> PATHOLOGICAL MATERIAL OF NO COMMERCIAL VALUE
> Department of Livestock Development
> Regional Reference Laboratory for FMD in South-East Asia
> Pak Chong, Nakhonratchasima, 30130
> THAILAND
> Tel: +66 44 279112
> Fax:+66 44 314889
> PERISHABLE FRAGILE KEEP AT 4 °C

4. Dispatch

All biological materials must be sent by airfreight direct to Suvarnabhumi International Airport. Before dispatching samples, the sender must notify the Regional Reference Laboratory at Pak Chong by facsimile (+66 44 314889). The institution submitting the samples should provide details of the air waybill number, flight number and time and date of arrival of the package in Bangkok. Staff of the Department of Livestock Development (DLD) and Regional Reference Laboratory will clear the parcel through customs at the airport. The parcel will be collected and taken to Pak Chong by staff of the Regional Reference Laboratory.

F.3 Samples for classical swine fever

The principles outlined above for FMD sample collection can be applied to other diseases, such as CSF. Specific details for CSF are as follows:

– For virus identification, the following samples should be collected during post-mortem examination: tonsil, lymph nodes (mesenteric and pharyngeal), kidney, and distal ileum.

– From live animals, blood should be collected into EDTA.

– For serological testing, serum samples should be taken from recovered animals, from sows with congenitally affected litters, and from pigs under surveillance.

– All samples should be refrigerated and shipped to the laboratory as soon as possible.

Note: Detailed information on sample collection and submission can be found in:

OIE (2017). Terrestrial Animal Health Code. World Organisation for Animal Health. Available at: http://www.oie.int/en/international-standard-setting/terrestrial-code/access-online/

OIE (2017). Manual of Diagnostic Tests and Vaccines for Terrestrial Animals. Available at: http://www.oie.int/en/international-standard-setting/terrestrial-manual/access-online/

OIE Technical disease cards: http://www.oie.int/animal-health-in-the-world/technical-disease-cards/

G. Participatory epidemiology

G.1 Key principles of participatory appraisal

– Behaviour and attitude
  - Listen, learn and respect
  - Be prepared to unlearn negative attitudes and stereotypes
  - Act as a facilitator, not an expert
– People are knowledgeable
  - On subjects important to their livelihoods
  - Certain individuals have unique and valuable perspectives
– Co-learning
  - Share knowledge, experience and analysis
  - Combine local and professional knowledge for effective acceptable action
– People are rational
  - There is an insider's and an outsider's perception of behaviour
  - Based on the information available, most people make rational decisions
  - The appearance of irrational behaviour means that a misunderstanding may have occurred
– Optimal knowledge/optimal ignorance
  - There is a balance between the need for information and the need for timely decision-making
– Action-orientated
  - Be prepared to take action rather than just collect data

G.2 Summary guidelines for semi-structured interviews

1. Prepare yourself: this is possibly the most important stage! Define the topic you want to investigate, work out the key 4 or 5 questions you want to ask and who it is you want to interview. If possible, bring an assistant along as a note-taker.

2. Introduce yourself and the purpose of the meeting: Your informants will want to know why you have come and why you have an interest in the selected topic.

3. Watch your body language throughout: Be friendly, informal and respectful, and try to sit on the ground! Stay calm: there is never any need to become emotional!

4. Start with general questions/comments: This will put people at ease. The easiest way to begin is to start with something visible that everybody can agree with. Use simple language. Ask only one question at a time.

5. Mix questions with general discussion: By introducing variety, you will keep up the interest of your informants. Casual dialogue will ensure good communication.

6. Use diagrams, symbols, and other drawings: These will help in keeping people interested and ensuring everybody participates and understands.

7. Use simple language: Avoid 'scientific' words. Ask only one question at a time, avoid leading questions, long or complicated questions, or questions that can be answered with a simple 'yes' or 'no'.

8. Probe: This is the most difficult stage. If an interesting point comes up, try to discover more. Six small words (why, how, who, what, when, where?) will help you to probe: keep them in mind throughout!

9. Observe: Watch closely to make sure that everybody participates (especially women) and the conversation is not dominated by a few individuals. Also make sure that people are not getting restless (a sign that they are getting tired): normally, 90 minutes is the maximum for group interviews.

10. Thank the participants: When the interview is over, thank your informants and give them an opportunity to ask their own questions: this is polite and will give you valuable clues!

11. Make full notes *after* the interview (unless you have a note taker): Wait until the interview is over before you write full notes. During the interview itself, only write down the main points so that you do not slow down or interrupt the conversation.

G.3 Summary guidelines for focus group discussions (FGD)

1. Determine the purpose: have clear objectives

2. Analyse the situation: build good knowledge of local conditions or start with interviews with key informants

3. Prepare a discussion guide: it has to be a written guide with open-ended questions. Avoid 'why' questions and those that require only a yes/no answer. List questions in logical order, from general to more specific, from factual to more sensitive.

4. Choose the right group: you can choose heterogeneous or homogenous groups. Homogenous groups (similar age, sex, socio-economic background) are often preferred, because it often facilitates free discussions; however, to obtain a wide range of views quickly, you will often need to interview a heterogenous group with a mix of participants (old/young, urban/rural, poor/not poor, etc.).

5. Select the right participants: pick participants that are likely to express a range of views. To get the right variety of participants, the choice should be random from a group. If there is no good way of finding out suitable persons try avoiding convenience sample (e.g. people easily accessible).

6. Arrange the physical environment: choose a 'neutral', easily accessible place and seat everyone in a circle. Avoid disturbances from other people, have adequate light and sufficient silence, and provide drinks and/or snacks (not noisy).

7. Conduct the session appropriately: one moderator will conduct the session. He/she should not act as an expert, but stimulate and support discussions. If the moderator is an 'expert', questions may be answered after the session. One note-taker/recorder will record what is happening in the group, e.g. reactions, feelings and comments from participants. A tape-recorder is generally used.

8. Obtain consent from participants: the moderator and note-taker/recorder should introduce themselves, give the participants information about the topic of discussion and explain how the session will proceed. They must then obtain consent from participants before the session begins and ask for permission to use a tape-recorder.

9. Encourage discussion: create an unthreatening environment. Moderate, listen, observe, and analyse. Show interest and be careful not to make judgements, there are no right or wrong answers. Encourage involvement of the participants. Use, "Can you tell me more about…", "What about you?", "What is your view…? Try to deal with 'Dominating' and 'Reluctant' persons.

10. Build rapport and empathise: observe non-verbal communication and handle sensitive issues (write down the participants' comments anonymously). Control the timing, but in a subtle way (allocated time to various topics), to maintain interest. At the end, summarise, check for agreement and ask for additional comments.

Considerations to make before running FGD and when analysing responses:

1. People often do not understand why they are doing the things they are doing, and therefore cannot tell you.

2. Sometimes people are in touch with their reasons and their feelings, but they cannot express them.

3. Even when they do understand why they are doing things, they often do not want to tell you.

4. When they do tell you, they often do not tell you the truth, or the whole truth.

5. Often people are not interested in examining their motivations.

There is rarely a single reason why a given person does something.

G.4 Method for drawing an outbreak risk map

Imagine, for example, you are conducting a study on CSF in pigs, and you are interested in identifying risk factors for the introduction of CSF to village pig populations and risk factors for CSF spread between different pig populations. You have travelled to a village that recently experienced an outbreak of CSF in its pigs to study the outbreak and identify these risk factors.

1. Begin by creating a participatory map with the villagers, making sure that all the geographic and physical features are included.

2. Discuss with the villagers what animal diseases are important to them. Since they recently had an outbreak of CSF in their pigs they will likely mention CSF, and then you can focus on your disease of interest from that point forward.

3. Work through the various disease determinants that you are familiar with. You might have them listed, or you might use a mental checklist. To name a few: first household affected, second household affected, spread of the disease through the village, types of pigs affected, nearest pig market, homes of pig traders, movement patterns of pig traders on bicycles or motorcycles, location of commercial pig farmers, movement patterns of suppliers (feed, litter, piglets, etc.) and buyers (piglets, weaners, finishers, meat, etc.) for commercial operations, location of butchers, location of rivers and gardens, houses that have pig houses, places where free-range pigs like to gather, etc.

4. Ask probing questions as details develop on the map. For example, once the pig market and the first affected house are mapped, you can ask: When was the last time people in this house visited the pig market? Did they bring back any new pigs and put them their pen?

5. Be sure to observe closely. When the path of the pig trader and the path of the outbreak spread are drawn, look to see if they are similar. If they are, ask questions to probe deeper. For example: When did the outbreak start? When did the pig trader last pass through the village?

6. Keep your mind open to discovering new risk factors as the map and the discussion develops. Do not simply run through the risk factors that you are already familiar with.

7. Remember that there may be sensitivities about an outbreak. Be careful not to create an atmosphere of recrimination, where some people begin blaming others for their disease problems. For example, the informants may want to hold the person in the first affected household responsible if they begin to feel that the outbreak was their fault. Make sure the atmosphere of the interview is one of open discovery, so that what is learned can be used by both you and the members of the villagers to prevent outbreaks in the future, not to exact retribution for outbreaks in the past.

8. When the exercise is complete, discuss with the villagers what they have learned, and how this may help them prevent outbreaks in the future.

G.5 Proportional piling method

1. Have your ranking criteria clear in your own mind.

2. Use open-ended questions to develop the list of items or categories for scoring. For example, 'what are the health problems that affected cattle and buffalo in the community in the last year?'

3. Probe the responses. Ask for descriptions and clarifications.

4. Explain that you want to carry out an exercise to better understand what you are learning about their health problems.

Draw circles on the ground, one circle for each disease mentioned, and place a drawing or card next to each circle that illustrates the disease. Circles can also be made from construction paper or drawn on flipchart paper.

5. Make sure that everyone recognises each category by its drawing or card.

6. Place 100 counters in a pile, and ask the respondents to divide them according to a particular characteristic or parameter. Respondents should not count the counters, but divide them visually. Record the question now if you have not already. For example, 'Please divide these beans to represent the impact each disease had on your livelihood in the past year'.

7. Allow time for discussion to enable respondents to reach a consensus about how to divide the piles. When the group appears to be finished, summarise and crosscheck the result. "Does everyone agree? Does anyone disagree that tuberculosis has such a big impact?"

8. Count the counters, but leave them in place so that the result can be discussed.

9. Probe the results. Why did they make these choices?

Example of proportional piling method to estimate disease incidence. Using a pile of 100 stones, the informant was asked to divide the stones to show the pattern of 'sick cattle during the last year' and 'healthy cattle during the last year'. The pile of stones representing sick cattle was then sub-divided by the informant to show the pattern of cattle having each of the main five diseases previously identified by the informant through semi-structure interviews, plus a category called 'other diseases' (a total of six disease categories). Each pile of stones was then further sub-divided to show the pattern of cattle dying and surviving for each disease category. This resulted in two piles of stones for each of the five diseases and the 'other diseases' category (Catley et al., 2004).

G.6 Matrix scoring disease definition method

Imagine that you would like to conduct a matrix scoring exercise to understand the clinical signs a community associates with different diseases.

1. Use the diseases mentioned by your respondents. When your respondents tell you the name of a disease in their language, use that name during the interview rather than an English or scientific name. That way, everybody, respondents and research team alike, is on the same page.

2. Obtain a list of clinical signs and epidemiological features for each disease.

3. Draw a matrix on the ground or on flipchart paper. Make sure it is big enough for everyone to see it. Include a column for each of the diseases. Use pictures, objects, or cards to represent the diseases and place these across the top of the matrix. Be sure to mention which disease each card represents, using the local language name, as you place it on the ground. This way, those that cannot read or understand the picture can memorise the cards as they appear.

4. Use all the indicators (clinical signs) mentioned by the respondents for the various diseases. Write the first indicator on a card, or use a picture or object to represent it. Place this to one side of the first row of the matrix. Be sure to repeat it aloud so that all the participants know what it is.

5. Place a pile of 30 counters next to the indicator and ask the participants to use them to show how strongly the indicator correlates with each disease. Summarise and crosscheck for agreement between the respondents. Leave the counters where they are.

6. Repeat the exercise for each indicator one by one, gradually building up the matrix. Leave the matrix in place so that everyone can view the results and discuss as a group.

7. Carefully probe the informants as to why they are scoring the way they are, both during the exercise and after the matrix is complete. Finally, summarise the results and give the informants the opportunity to make changes if they wish. Ask your respondents what new learning or insights they have gained from the exercise.

8. Record the results in a matrix in your notebook.

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