Annex XVII

CHAPTER 12.1.

INFECTION WITH AFRICAN HORSE SICKNESS VIRUS

Article 12.1.1.

General provisions

For the purposes of the *Terrestrial Code*, African horse sickness (AHS) is defined as an *infection* of equids with African horse sickness virus (AHSV).

The following defines an *infection* with AHSV:

1) AHSV has been isolated and identified from an equid or a product derived from that equid; or

2) viral antigen or viral ribonucleic acid ~~(RNA)~~ specific to ~~a serotype of~~ AHSV has been identified in samples from an equid showing clinical signs consistent with AHS, or epidemiologically linked to a suspected or confirmed *infection* ~~*case*~~; or

3) serological evidence of active *infection* with AHSV by detection of seroconversion with production of antibodies against structural or nonstructural proteins of AHSV that are not a consequence of *vaccination* have been identified in an equid that either shows clinical signs consistent with AHS, or is epidemiologically linked to a suspected or confirmed *infection* ~~*case*~~.

For the purposes of the *Terrestrial Code*, the *infective period* for ~~African horse sickness virus~~ (AHSV) shall be 40 days for domestic horses. Although critical information is lacking for some species, this chapter applies to all equidae.

All countries or *zones* adjacent to a country or *zone* not having free status should determine their AHSV status from an ongoing *surveillance* programme. Throughout the chapter, *surveillance* is in all cases understood as being conducted as described in Article 12.1.~~13~~11. to 12.1.~~15~~13.

~~The following defines a case of African horse sickness (AHS):~~

~~1) AHSV has been isolated and identified from an equid or a product derived from that equid; or~~

~~2) viral antigen or viral RNA specific to one or more of the serotypes of AHSV has been identified in samples from one or more equids showing clinical signs consistent with AHS, or epidemiologically linked to a suspected or confirmed *case*; or~~

~~3) serological evidence of active infection with AHSV by detection of seroconversion with production of antibodies to structural or nonstructural proteins of AHSV that are not a consequence of *vaccination* have been identified in one or more equids that either show clinical signs consistent with AHS, or epidemiologically linked to a suspected or confirmed *case*.~~

Standards for diagnostic tests and vaccines are described in the *Terrestrial Manual*.

Article 12.1.2.

~~AHSV free~~ Country or zone free from infection with AHSV

1) A country or *zone* may be considered free from *infection* with AHSV when ~~African horse sickness (~~*infection* with ~~AHS~~V~~)~~ is notifiable in the whole country, systematic *vaccination* is prohibited, importation of equids and their semen, oocytes or embryos are carried out in accordance with this chapter, and either:

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a) historical freedom as described in Chapter 1.4. has demonstrated no evidence of AHSV in the country or *zone*; or

b) the country or *zone* has not reported any *case* of AHSV *infection* for at least two years and is not adjacent to an infected country or *zone*; or

c) a *surveillance* programme has demonstrated no evidence of AHSV in the country or *zone* for at least ~~twenty-four months~~ two years; or

d) the country or *zone* has not reported any ~~*case of*~~ AHSV *infection* for at least 40 days and a *surveillance* programme has demonstrated no evidence of *Culicoides* for at least two years in the country or *zone*.

2) ~~An AHS free~~ Country or *zone* free from *infection* with AHSV which is adjacent to an infected country or *infected zone* should include a *zone* in which *surveillance* is conducted in accordance with Articles 12.1.~~1311~~. to 12.1.~~1513~~. ~~*Animals* within this *zone* should be subjected to continuing *surveillance*. The boundaries of this *zone* should be clearly defined, and should take account of geographical and epidemiological factors that are relevant to AHS transmission.~~

3) A~~n AHSV free~~ country or *zone* free from *infection* with AHSV will not lose its free status through the importation of ~~vaccinated or~~ seropositive or vaccinated equids and their semen, oocytes or embryos from infected countries or *infected zones*, provided these imports are carried out in accordance with this chapter.

4) To qualify for inclusion in the list of AHSV free countries or *zones*, a Member Country should:

a) have a record of regular and prompt animal disease reporting;

b) send a declaration to the OIE stating:

i) the section under point 1) on which the application is based;

ii) no routine *vaccination* against AHS has been carried out during the past ~~twelve months~~ year in the country or *zone*;

iii) equids are imported in accordance with this chapter;

c) supply documented evidence that:

i) *surveillance* in accordance with Articles 12.1.~~1311~~. to 12.1.~~1513~~. is applied, unless historically free;

ii) regulatory measures for the early detection, prevention and control of *infection* with AHSV have been implemented.

5) The Member Country will be included in the list only after the submitted evidence has been accepted by the OIE. Retention on the list requires that the information in points 4b)ii) and iii) and 4c) ~~ii)~~ above be re-submitted annually and changes in the epidemiological situation or other significant events be reported to the OIE according to the requirements in Chapter 1.1., and in particular, formally state that :

a) there has been no *outbreak* of AHS during the past ~~twelve months~~ year in the country or *zone*;

b) no evidence of *infection* with AHSV ~~infection~~ has been found during the past ~~twelve months~~ year in the country or *zone*.

~~Article 12.1.3.~~

~~**AHSV seasonally free zone**~~

~~1) An AHSV seasonally free *zone* is a part of an infected country or an *infected zone* in which for part of a year, ongoing *surveillance* and monitoring consistently demonstrated neither evidence of AHSV transmission nor the evidence of the presence of adult *Culicoides*.~~

~~2) AHS is notifiable in the whole country.~~

~~3)~~ ~~For the application of Articles 12.1.8., 12.1.10. and 12.1.11., the seasonally free period is:~~

~~a)~~ ~~taken to commence the day following the last evidence of AHSV transmission and of the cessation of activity of adult *Culicoides* as demonstrated by an ongoing *surveillance* programme, and~~

~~b)~~ ~~taken to conclude either:~~

~~i)~~ ~~at least 40 days before the earliest date that historical data show AHSV activity has recommenced; or~~

~~ii)~~ ~~immediately when current climatic data or data from a *surveillance* and monitoring programme indicate an earlier resurgence of activity of adult *Culicoides vectors*.~~

~~4.~~ ~~An AHSV seasonally free *zone* will not lose its free status through the importation of vaccinated or seropositive equids and their semen, oocytes or embryos from infected countries or *infected zones*, provided these imports are carried out in accordance with this chapter.~~

Article 12.1.~~4~~3.

AHSV infected country or zone

For the purpose of this chapter, an AHSV infected country or *zone* is one that does not fulfil the requirements to qualify as ~~either AHSV free country or *zone*~~ free from *infection* with AHSV ~~or AHSV seasonally free *zone*~~.

Article 12.1.~~5~~4.

Establishment of a containment zone within a~~n~~ ~~AHS free~~ country or zone free from infection with AHSV

In the event of limited *outbreaks* within a~~n~~ ~~AHS free~~ country or zone free from *infection* with AHSV, ~~including within a *protection zone*,~~ a single *containment zone*~~, which includes all *cases*~~, ~~and should be large enough to contain any potentially infected vectors,~~ can be established for the purpose of minimising the impact on the entire country or *zone*. Such a *zone* should include all *cases* and can be established within a *protection zone*. For this to be achieved, the *Veterinary Authority* should provide documented evidence that:

1) the *outbreaks* are limited based on the following factors:

   a) immediately on suspicion, a rapid response including notification has been made;

   b) standstill of movements of equids has been imposed, and effective controls on the movement of equids and their products specified in this chapter are in place;

   c) epidemiological investigation (trace-back, trace-forward) has been completed;

   d) the *infection* has been confirmed;

   e) ~~the primary *outbreak* and likely source of the *outbreak* has been identified;~~ investigations on the likely source of the *outbreak* have been carried out;

   f) all *cases* have been shown to be epidemiologically linked;

   g) no new *cases* have been found in the *containment zone* within a minimum of two *infective periods* as defined in Article 12.1.1.;

2) the equids within the *containment zone* ~~should be~~ are clearly identifiable as belonging to the *containment zone*;

3) increased passive and targeted *surveillance* in accordance with Articles 12.1.~~13~~11. to 12.1.~~15~~13. in the rest of the country or *zone* has not detected any evidence of *infection*;

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4) animal health measures are in place to ~~that~~ effectively prevent the spread of AHSV *infection* to the rest of the country or *zone*, taking into consideration the establishment of a *protection zone* within the *containment zone*, the seasonal vector conditions and existing physical, geographical and ecological barriers;

5) ongoing *surveillance* in accordance with Articles 12.1.~~13~~11. to 12.1.~~15~~13. is in place in the *containment zone*.

The free status of the areas outside the *containment zone* is suspended ~~pending the establishment of~~ while the *containment zone* is being established in accordance with points 1 to 5 above. The free status of the areas outside the *containment zone* ~~could~~ may be reinstated irrespective of the provisions of Article 12.1.~~6~~5., once the *containment zone* is recognised by the OIE.

In the event of the recurrence of AHSV in the *containment zone*, the approval of the *containment zone* is withdrawn.

The recovery of the AHSV free status of the *containment zone* should follow the provisions of Article 12.1.~~6~~5.

Article 12.1.~~6~~5.

Recovery of free status

To regain the free status when an AHS *outbreak* occurs in a~~n AHS free~~ country or *zone* previously free from infection with AHSV, ~~to regain the free status,~~ the provisions of Article 12.1.2. apply, irrespective of whether emergency *vaccination* has been applied.

Article 12.1.~~7~~6.

Recommendations for importation from ~~AHSV free~~ countries or zones free from infection with AHSV

For equids

Veterinary Authorities should require the presentation of an *international veterinary certificate* attesting that the animals:

1) showed no clinical sign of AHS on the day of shipment;

2) have not been vaccinated against AHS within the last 40 days;

3) were kept in ~~an AHSV free~~ country~~(~~ies~~)~~ or *zone*~~(~~s~~)~~ free from infection with AHSV since birth or for at least 40 days prior to shipment;

4) either:

   a) did not transit through an *infected zone* during transportation to the *place of shipment*; or

   b) were protected from attacks from *Culicoides* at all times when transiting through an *infected zone*.

~~Article 12.1.8.~~

~~**Recommendations for importation from AHSV seasonally free zones during the seasonally free period**~~

~~For equids~~

~~*Veterinary Authorities* should require the presentation of an *international veterinary certificate* attesting that the animals:~~

~~1) showed no clinical signs of AHS on the day of shipment;~~

~~2) have not been vaccinated against AHS within the last 40 days;~~

~~3) and either~~

~~a) were kept in an AHSV seasonally free *zone* during the seasonally free period since birth or for at least 40 days prior to shipment; or~~

~~b) were held in isolation in a *vector*-protected *establishment* prior to shipment~~

~~i) for a period of at least 28 days and a serological test according to the *Terrestrial Manual* to detect antibodies to the AHSV group, was carried out with a negative result on a blood sample collected at least 28 days after introduction into the *vector* protected *establishment*; or~~

~~ii) for a period of at least 40 days and serological tests according to the *Terrestrial Manual* to detect antibodies against AHSV were carried out with no significant increase in antibody titre on blood samples collected on two occasions, with an interval of not less than 21 days, the first sample being collected at least seven days after introduction into the *vector* protected *establishment*; or~~

~~iii) for a period of at least 14 days and an agent identification tests according to the *Terrestrial Manual* was carried out with a negative results on a blood samples collected not less than 14 days after introduction into the *vector* protected *establishment*;~~

~~4) were protected from attacks from *Culicoides* at all times when transiting through an *infected zone*.~~

Article 12.1.~~9~~7.

Recommendations for importation from AHSV infected countries or zones

For equids

Veterinary Authorities should require the presentation of an *international veterinary certificate* attesting that the animals:

1) showed no clinical sign of AHS on the day of shipment;

2) have not been vaccinated against AHS within the last 40 days;

3) were held in isolation in a *vector*-protected *establishment*

a) for a period of at least 28 days and a serological test ~~according to the *Terrestrial Manual*~~ to detect antibodies to the AHSV group, was carried out with a negative result on a blood sample collected at least 28 days after introduction into the *vector*-protected *establishment*; or

b) for a period of at least 40 days and serological tests ~~according to the *Terrestrial Manual*~~ to detect antibodies against AHSV were carried out with no significant increase in antibody titre on blood samples collected on two occasions, with an interval of not less than 21 days, the first sample being collected at least seven days after introduction into the *vector*-protected *establishment*; or

c) for a period of at least 14 days and an agent identification test~~s~~ ~~according to the *Terrestrial Manual*~~ was carried out with a negative result~~s~~ on a blood sample~~s~~ collected not less than 14 days after introduction into the *vector*-protected *establishment*; or

d) for a period of at least 40 days and were vaccinated, at least 40 days before shipment, ~~in accordance with the *Terrestrial Manual*~~ against all serotypes whose presence in the source population has been

demonstrated through a *surveillance* programme in accordance with Articles 12.1.~~14~~12. and 12.1.~~15~~13, and were identified in the accompanying certification as having been vaccinated;

4) were protected from attacks by *Culicoides* at all times during transportation, ~~(~~including transportation to and at the *place of shipment*~~)~~.

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Article 12.1.~~10~~8.

Recommendations for the importation of equine semen

Veterinary Authorities of *importing countries* should require the presentation of an *international veterinary certificate* attesting that the donor animals:

1) showed no clinical sign of AHS on the day of collection of the semen and for the following 40 days;

2) had not been immunised against AHS with a live attenuated vaccine within 40 days prior to the day of collection;

3) were either:

   a) kept in a~~n AHSV free~~ country or ~~free~~ *zone* free from infection with AHSV ~~or from an AHSV seasonally free *zone* (during the seasonally free period)~~ for at least 40 days before commencement of, and during collection of, the semen, or

   b) kept in an AHSV free vector-protected *artificial insemination centre* throughout the collection period, and subjected to either:

      i) a serological test ~~according to the *Terrestrial Manual*~~ to detect antibody to the AHSV group, carried out with a negative result on a blood sample collected at least 28 days and not more than 90 days after the last collection of semen; or

      ii) agent identification tests ~~according to the *Terrestrial Manual*~~ carried out with negative results on blood samples collected at commencement and conclusion of, and at least every seven days, during semen collection for this consignment.

Article 12.1.~~11~~9.

Recommendations for the importation of *in vivo* derived equine embryos or oocytes

Veterinary Authorities of *importing countries* should require the presentation of an *international veterinary certificate* attesting that:

1) the donor animals:

   a) showed no clinical sign of AHS on the day of collection of the embryos or oocytes and for the following 40 days;

   b) had not been immunised against AHS with a live attenuated vaccine within 40 days prior to the day of collection;

   c) were either:

      i) kept in a~~n AHSV free~~ country or ~~free~~ *zone* free from infection with AHSV ~~or from an AHSV seasonally free *zone* (during the seasonally free period)~~ for at least 40 days before commencement of, and during collection of the embryos or oocytes, or

      ii) kept in an AHSV free vector-protected *collection centre* throughout the collection period, and subjected to either:

         - a serological test ~~according to the *Terrestrial Manual*~~ to detect antibody to the AHSV group carried out with a negative result on a blood sample collected at least 28 days and not more than 90 days after the last collection of embryos or oocytes; or

- agent identification tests ~~according to the *Terrestrial Manual*~~ carried out with negative results on blood samples collected at commencement and conclusion of, and at least every seven days during embryos or oocytes collection for this consignment;

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2) the embryos were collected, processed and stored in conformity with the provisions of Chapter 4.7. or Chapter 4.9., as relevant;

3) semen used to fertilise the oocytes, complies at least with the requirements in Article 12.1.~~10~~8.

Article 12.1.~~12~~10.

Protecting animals from *Culicoides* attack

1. Vector-protected establishment or facility

The *establishment* or facility should be approved by the *Veterinary Authority* and the means of protection should at least comprise the following;

a) appropriate physical barriers at entry and exit points, for example double-door entry-exit system;

b) openings of the building are *vector* screened with mesh of appropriate gauge impregnated regularly with an approved insecticide according to manufacturers' instruction;

c) *vector surveillance* and control within and around the building;

d) measures to limit or eliminate breeding sites for *vectors* in vicinity of the *establishment* or facility;

e) Standard Operating Procedure, including description of back-up and alarm systems, for operation of the *establishment* or facility and transport of ~~horses~~ equids to the place of *loading*.

2. During transportation

When transporting equids through AHSV infected countries or AHSV *infected zones*, *Veterinary Authorities* should require strategies to protect animals from attacks by *Culicoides* during transport, taking into account the local ecology of the *vector*.

a) Transport by road:

Potential *risk management* strategies include a combination of:

i) treating animals with chemical repellents prior to and during transportation, in sanitized *vehicles* treated with appropriate residual contact insecticide;

ii) *loading*, transporting and *unloading* animals at times of low *vector* activity (i.e. bright sunshine and low temperature);

iii) ensuring *vehicles* do not stop en route during dawn or dusk, or overnight, unless the animals are held behind insect proof netting;

iv) darkening the interior of the *vehicle*, for example by covering the roof or sides of *vehicles* with shade cloth;

v) ~~*monitoring*~~ *surveillance* for *vectors* at common stopping and offloading points to gain information on seasonal variations;

vi) using historical, ongoing or ~~AHS~~ modelling information on AHS to identify low risk ports and transport routes.

b) Transport by air:

Prior to *loading* the equids, the crates, *containers* or jet stalls are sprayed with an insecticide approved in the country of dispatch.

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Crates, *containers* or jet stalls in which equids are being transported and the cargo hold of the aircraft should ~~must~~ be sprayed with an approved insecticide when the doors have been ~~just after the doors to the aircraft are~~ closed and prior to take-off, ~~or immediately prior to the closing of the aircraft doors after loading~~. All possible insect harbourage should be treated. The spray containers should be retained for inspection on arrival.

In addition, during any stopover in countries or *zones* not free ~~of~~ from infection with AHSV, prior to~~, or immediately after~~ the opening of any aircraft door and until all doors are closed, netting of appropriate gauge impregnated with an approved insecticide should ~~must~~ be placed over all crates, *containers* or jet stalls.

Article 12.1.~~13~~11.

Introduction to surveillance~~: introduction~~

Articles 12.1.~~13~~11. to 12.1.~~15~~13. define the principles and provide guidance on *surveillance* for AHSV *infection*, complementary to Chapter 1.4. and, for *vectors*, complementary to Chapter 1.5.

AHS is a *vector*-borne *infection* transmitted by a limited number of species of *Culicoides* insects. Unlike the related bluetongue virus, AHSV is so far geographically restricted to sub Saharan Africa with periodic excursions into North Africa, southwest Europe, the Middle East and adjacent regions of Asia. An important component of AHSV epidemiology is vectorial capacity which provides a measure of *disease risk* that incorporates *vector* competence, abundance, seasonal incidence, biting rates, survival rates and the extrinsic *incubation period*. However, methods and tools for measuring some of these *vector* factors remain to be developed, particularly in a field context.

According to this chapter, a Member Country demonstrating freedom from AHSV infection for the entire country or a *zone* should provide evidence for the existence of an effective *surveillance* programme. The strategy and design of the *surveillance* programme will depend on the prevailing epidemiological circumstances and should be planned and implemented according to general conditions and methods described in this chapter. This requires the support of a *laboratory* able to undertake identification of AHSV infection through the virus detection and antibody tests ~~described in the *Terrestrial Manual*~~.

Susceptible *captive wild, feral and wild* equine populations should be included in the *surveillance* programme.

~~For the purposes of *surveillance*, a case refers to an equid infected with AHSV.~~

The purpose of *surveillance* is to determine if a country or *zone* is free from infection with AHSV ~~or if a *zone* is seasonally free from AHSV~~. *Surveillance* deals not only with the occurrence of clinical signs caused by AHSV, but also with evidence of infection with AHSV in the absence of clinical signs.

Article 12.1.~~14~~12.

~~Surveillance:~~ General surveillance conditions and methods

1) A *surveillance* system should be under the responsibility of the *Veterinary Authority*. In particular the following should be in place:

   a) a formal and ongoing system for detecting and investigating *outbreaks* of *disease*;

   b) a procedure for the rapid collection and transport of samples from suspected *cases* of AHSV *infection* to a *laboratory* for ~~AHS~~ diagnosis ~~as described in the *Terrestrial Manual*~~;

   c) a system for recording, managing and analysing diagnostic, epidemiologic and *surveillance* data.

2) The AHSV *infection* *surveillance* programme should:

   a) in a ~~country/ *zone*~~ free ~~or seasonally free~~ country or *zone*, have ~~include~~ an early warning system which oblige ~~for reporting suspicious *cases*.~~ Ppersons who have regular contact with equids, as well as diagnosticians, ~~should~~ to report promptly any suspicion of AHSV infection to the *Veterinary Authority*. An effective *surveillance* system will periodically identify ~~suspicious~~ suspected *cases* that require follow-up and investigation to confirm or exclude that the cause of the condition is AHS. The rate at which such ~~suspicious~~ suspected *cases* are likely to occur will differ between epidemiological situations and cannot therefore be predicted reliably. All suspected *cases* of AHSV *infection* should be investigated immediately and samples should be taken and submitted to a *laboratory*. This requires that sampling kits and other equipment ~~are~~ be available ~~for~~ to those responsible for *surveillance*;

b) conduct random or targeted serological and virological *surveillance* appropriate to the *infection* status of the country or *zone* in accordance with Chapter 1.4.

Article 12.1.~~15~~13.

Surveillance strategies

The target population for *surveillance* aimed at identification of *disease* or *infection* should cover susceptible equids within the country or *zone*. Active and passive *surveillance* for AHSV infection should be ongoing. *Surveillance* should be composed of random or targeted approaches using virological, serological and clinical methods appropriate for the *infection* status of the country or *zone*.

A Member Country should justify the *surveillance* strategy chosen as appropriate to detect the presence of AHSV infection in accordance with Chapter 1.4. and the prevailing epidemiological situation. It may, for example, be appropriate to target clinical *surveillance* at particular species likely to exhibit clinical signs (e.g. horses). Similarly, virological and serological testing may be targeted to species that rarely show clinical signs (e.g. donkeys).

In vaccinated populations serological and virological *surveillance* is necessary to detect the AHSV types circulating to ensure that all circulating types are included in the *vaccination* programme.

If a Member Country wishes to declare freedom from AHSV infection in a specific *zone*, the design of the *surveillance* strategy would need to be aimed at the population within the *zone*.

For random surveys, the design of the sampling strategy will need to incorporate epidemiologically appropriate design prevalence. The sample size selected for testing will need to be large enough to detect *infection* if it were to occur at a predetermined minimum rate. The sample size, expected prevalence and diagnostic sensitivity of the tests determine the level of confidence in the results of the survey. The Member Country must justify the choice of design prevalence and confidence level based on the objectives of *surveillance* and the epidemiological situation, in accordance with Chapter 1.4. Selection of the design prevalence, in particular, needs to be based on the prevailing or historical epidemiological situation.

Irrespective of the survey approach selected, the sensitivity and specificity of the diagnostic tests employed are key factors in the design, sample size determination and interpretation of the results obtained. Ideally, the sensitivity and specificity of the tests used should be validated for the *vaccination* or *infection* history and the different species in the target population.

Irrespective of the testing system employed, *surveillance* system design should anticipate the occurrence of false positive reactions. If the characteristics of the testing system are known, the rate at which these false positives are likely to occur can be calculated in advance. There needs to be an effective procedure for following up positives to ultimately determine with a high level of confidence, whether they are indicative of *infection* or not. This should involve both supplementary tests and follow-up investigation to collect diagnostic material from the original sampling unit as well as those which may be epidemiologically linked to it.

The principles for *surveillance* for *disease* or *infection* are technically well defined. *Surveillance* programmes to prove the absence of AHSV infection or ~~circulation~~ transmission, need to be carefully designed to avoid producing results that are either insufficiently reliable to be accepted by international trading partners, or excessively costly and logistically complicated. The design of any *surveillance* programme, therefore, requires inputs from professionals competent and experienced in this field.

1. Clinical surveillance

Clinical *surveillance* aims at the detection of clinical signs of AHS in equids particularly during a newly introduced *infection*. In horses, clinical signs may include pyrexia, oedema, hyperaemia of mucosal membranes and dyspnoea.

AHS suspects detected by clinical *surveillance* should always be confirmed by *laboratory* testing.

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2. Serological surveillance

Serological *surveillance* of equine populations is an important tool to confirm absence of AHSV transmission in a country or *zone*. The species tested should reflect the local epidemiology of AHSV infection, and the equine species available. Management variables that may reduce the likelihood of *infection*, such as the use of insecticides and animal housing, should be taken into account when selecting equids to be included in the *surveillance* system.

Samples should be examined for antibodies against AHSV ~~using tests prescribed in the *Terrestrial Manual*~~. Positive AHSV antibody tests results can have four possible causes:

a) natural *infection* with AHSV;

b) *vaccination* against AHSV;

c) maternal antibodies;

d) positive results due to the lack of specificity of the test.

It may be possible to use sera collected for other purposes for AHSV *surveillance*. However, the principles of survey design described in these recommendations and the requirements for a statistically valid survey for the presence of AHSV infection should not be compromised.

The results of random or targeted serological surveys are important in providing reliable evidence that no AHSV infection is present in a country or *zone*. It is, therefore, essential that the survey is thoroughly documented. It is critical to interpret the results in light of the movement history of the animals being sampled.

Serological *surveillance* in a free *zone* should target those areas that are at highest risk of AHSV transmission, based on the results of previous *surveillance* and other information. This will usually be towards the boundaries of the free *zone*. In view of the epidemiology of AHSV, either random or targeted sampling is suitable to select *herds* or animals for testing.

Serological *surveillance* in a free country or *zone* should be carried out over an appropriate distance from the border with an infected country or *infected zone*, based upon geography, climate, history of *infection* and other relevant factors. The *surveillance* should be carried out over a distance of at least a hundred kilometres from the border with that country or *zone*, but a lesser distance could be acceptable if there are relevant ecological or geographical features likely to interrupt the transmission of AHSV. A~~n AHSV free~~ country or *zone* free from *infection* with AHSV may be protected from an adjacent infected country or *infected zone* by a *protection zone*.

Serological *surveillance* in *infected zones* will identify changes in the boundary of the *zone*, and can also be used to identify the AHSV types circulating. In view of the epidemiology of AHSV infection, either random or targeted sampling is suitable.

3. Virological surveillance

Isolation and genetic analysis of AHSV from a proportion of infected animals is beneficial in terms of providing information on serotype and genetic characteristics of the viruses concerned.

Virological *surveillance* ~~using tests described in the *Terrestrial Manual*~~ can be conducted:

a) to identify virus ~~circulation~~ transmission in at risk populations;

b) to confirm clinically suspected *cases*;

c) to follow up positive serological results;

d) to better characterise the genotype of circulating virus in a country or *zone*.

4. Sentinel animals

Sentinel animals are a form of targeted *surveillance* with a prospective study design. They comprise groups of unexposed equids that are not vaccinated and are managed at fixed locations and observed and sampled regularly to detect new AHSV infections.

The primary purpose of a sentinel equid programme is to detect AHSV infections occurring at a particular place, for instance sentinel groups may be located on the boundaries of *infected zones* to detect changes in distribution of AHSV. In addition, sentinel equid programmes allow the timing and dynamics of *infections* to be observed.

A sentinel equid programme should use animals of known source and history of exposure, control management variables such as use of insecticides and animal housing (depending on the epidemiology of AHSV in the area under consideration), and be flexible in its design in terms of sampling frequency and choice of tests.

Care is necessary in choosing the sites for the sentinel groups. The aim is to maximise the chance of detecting AHSV activity at the geographical location for which the sentinel site acts as a sampling point. The effect of secondary factors that may influence events at each location, such as climate, may also be analysed. To avoid confounding factors sentinel groups should comprise animals selected to be of similar age and susceptibility to AHSV infection. The only feature distinguishing groups of sentinels should be their geographical location. Sera from sentinel animal programmes should be stored methodically in a serum bank to allow retrospective studies to be conducted in the event of new serotypes being isolated.

The frequency of sampling should reflect the equine species used and the reason for choosing the sampling site. In endemic areas virus isolation will allow monitoring of the serotypes and genotypes of AHSV circulating during each time period. The borders between infected and non infected areas can be defined by serological detection of *infection*. Monthly sampling intervals are frequently used. Sentinels in declared free *zones* add to confidence that AHSV infections are not occurring unobserved. Here sampling prior to and after the possible period of transmission is sufficient.

Definitive information on AHSV circulating in a country or *zone* is provided by isolation and identification of the viruses. If virus isolation is required sentinels should be sampled at sufficiently frequent intervals to ensure that some samples are collected during the period of viraemia.

5. Vector surveillance

AHSV is transmitted between equine hosts by species of *Culicoides* which vary across the world. It is therefore important to be able to identify potential *vector* species accurately although many such species are closely related and difficult to differentiate with certainty.

Vector surveillance is aimed at demonstrating the absence of vectors or defining high, medium and low-risk areas and local details of seasonality by determining the various species present in an area, their respective seasonal occurrence, and abundance. *Vector surveillance* has particular relevance to potential areas of spread. Long term *surveillance* can also be used to assess *vector* abatement measures, or to confirm continued absence of vectors.

The most effective way of gathering this information should take account of the biology and behavioural characteristics of the local *vector* species of *Culicoides* and may include the use of Onderstepoort-type light traps or similar, operated from dusk to dawn in locations adjacent to equids.

Vector surveillance should be based on scientific sampling techniques. The choice of the number and types of traps to be used in *vector surveillance* and the frequency of their use should take into account the size and ecological characteristics of the area to be surveyed.

The operation of *vector surveillance* sites at the same locations as sentinel animals is advisable.

The use of a *vector surveillance* system to detect the presence of circulating virus is not recommended as a routine procedure as the typically low *vector infection* rates mean that such detections can be rare. ~~Other~~ *Animal*-based *surveillance* strategies are preferred to detect virus ~~circulation~~ transmission.

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