# EPIZOOTIC HAEMORRHAGIC DISEASE



## *AETIOLOGY*

### *Classification of the causative agent*

Epizootic haemorrhagic disease (EHD) is caused by a virus of the family *Reoviridae*, genus *Orbivirus*; there are 8 or more serotypes and Ibaraki virus is a member of the EHD virus (EHDV) serogroup (serotype 2). EHDV demonstrates immunological cross reactivity with the Bluetongue virus group.

### *Resistance to physical and chemical action* (*adapted from Bluetongue virus*)

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| **Temperature:** | Extremely unstable at high temperatures. Inactivated by 50°C (122°F)/3 hours; 60°C (140°F)/15 minutes or 121°C (249.8°F) /15 minutes. |
| **pH:** | Sensitive to pH <6.0 and >8.0. |
| **Chemicals/Disinfectants:** | Non-enveloped virus and thus relatively resistant to lipid solvents like ether and chloroform. Readily inactivated by ß-propiolactone, 2% w/v glutaraldehyde, acids, alkalis (2% w/v sodium hydroxide), 2-3% w/v sodium hypochlorite, iodophores and phenolic compounds. |
| **Survival:** | Very stable in blood and tissue specimens at 20°C/68°F, 4°C/39.2°F, and –70°C/–94°F, but not at –20°C/–4°F. Resistant to ultraviolet and gamma irradiation due to its double-stranded RNA genome. |

## *EPIDEMIOLOGY*

- EHD was considered an emerging disease in cattle, and was added to OIE list of notifiable diseases in May 2008, following outbreaks in four Mediterranean countries
- Morbidity and mortality may be as high as 90% in white tailed deer; however, severity varies depending on the year and geographical location

### *Hosts*

- EHD can infect most wild and domestic ruminants
- Historically, EHD has been a disease of wild ruminants, particularly white-tailed deer in North America. White-tailed deer are mainly affected, with mule deer and pronghorn affected to a lesser extent
- It was reported as a clinical disease of cattle, and never of small ruminants
- A notable exception is Ibaraki virus, which caused an extensive outbreak of disease in cattle in Japan in 1959, and continues to cause cattle disease in the Far East
- Serological evidence of EHDV infection has been reported in many ruminant species, both wild and domestic, including sheep, cattle, various species of deer, elk, bison, mountain goats and ibex, as well as camels, llamas, rhinoceros, bears, yaks and marsupials
- Other wild ruminants, like black-tailed deer, red deer, wapiti, fallow deer, roe deer, elk, moose, and bighorn sheep may seroconvert
- True persistent infection of ruminants does not occur
- Sheep can be infected experimentally but rarely develop clinical signs, and goats do not seem to be susceptible to infection

### *Transmission*

- Virus is transmitted by biological vectors, usually biting midges of the genus *Culicoides*, after an external extrinsic period of 10–14 days
- In temperate regions infection is most common in the late summer and autumn during peak vector population, while infection occurs throughout the year in tropical regions

- As in Bluetongue infection, viraemia can be prolonged beyond 50 days, despite the presence of neutralising antibody, due to an intimate association between virus and erythrocytes. Infected deer can be viraemic for up to 60 days
- EHDV is not known to cause disease in humans under any conditions.

### *Sources of virus*

- Blood of viraemic animals
- Infection in ruminants is not contagious – biological vectors (*Culicoides* sp.) are required
- As the virus infects endothelium, all tissues of the body may be affected

### *Occurrence*

- As a vector-borne viral disease, the distribution of EHD is limited to the distribution of competent *Culicoides* vectors
- The EHDV has been isolated from wild and domestic ruminants and arthropods in North America, South America, Asia, Africa, Islands in the Indian ocean (La Reunion and Mayotte) and Australia. Also, more recently in countries surrounding the Mediterranean Basin including Morocco, Algeria, Tunisia, Israel, Jordan and Turkey
- Outbreaks generally coincide with the peak of vector population abundance, so most cases of EHD occur in the late summer and autumn

**For more recent, detailed information on the occurrence of this disease worldwide, see the OIE World Animal Health Information Database (WAHID) Interface [http://www.oie.int/wahis/public.php?page=home].**

## *DIAGNOSIS*

The incubation period for EHD is estimated at 2–10 days.

### *Clinical diagnosis*

The clinical signs of EHD manifest as haemorrhagic disease in deer, but domestic ruminants may be subclinically infected.

- Acute EHD in deer: Fever, weakness, inappetence, excessive salivation, facial oedema, hyperaemia of the conjunctivae and mucous membranes of the oral cavity, coronitis stomatitis, and excessive salivation
- In prolonged cases, oral ulcers on the dental pad, hard palate, and tongue may occur. Excessive bleeding occurs in fulminant disease: bloody diarrhoea, haematuria, dehydration, and death
- Acute outbreaks in cattle (similar to Bluetongue): fever, anorexia, reduced milk, swollen conjunctivae, redness and scaling of the nose and lips, nasal and ocular discharge, stomatitis, salivation, lameness, swelling of the tongue, oral/nasal erosions, and dyspnoea
- Ibaraki disease in cattle is characterized by fever, anorexia and difficulty swallowing
- Oedema, haemorrhages, erosions, and ulcerations may be seen in the mouth, on the lips, and around the coronets; the animals may be stiff and lame
- Abortions and stillbirths have also been reported in some epidemics. Some affected cattle die (up to 10%)

### *Lesions*

<u>EHD in deer</u>:

- **Peracute form**: Severe oedema of the head, neck, tongue, conjunctiva, and lungs
- **Acute form**: widespread haemorrhages and oedema in the mucous membranes, skin and viscera, especially heart and gastrointestinal tract
    - Erosions may be found in the mouth, rumen and omasum, and necrosis in the hard palate, tongue, dental pads, oesophagus, larynx, rumen and abomasum
- **Chronic form**: growth rings on the hooves or sloughing of the hoof wall, and erosions, ulcers or scars in the rumen

Ibaraki disease in cattle:

- Degeneration of the striated muscles in the oesophagus, larynx, pharynx, tongue, and skeletal muscles with secondary aspiration pneumonia, dehydration and emaciation
- Marked oedema and haemorrhages in the mouth, lips, abomasum, and coronets may be observed. Erosions or ulcerations may also be present

## *Differential diagnosis*

- Deer: indistinguishable from Bluetongue, foot and mouth disease
- Cattle: Bluetongue, bovine viral diarrhoea, foot and mouth disease, infectious bovine rhinotracheitis, vesicular stomatitis, malignant catarrhal fever, and bovine ephemeral fever

## *Laboratory diagnosis*

**Samples**

*Virus isolation and detection*

- Whole blood in EDTA and/or heparin (virus isolation), in EDTA or citrate (PCR)
- Spleen
- Lungs
- Lymph nodes
- Liver

*Serological tests*

- Paired serum samples (3–5 ml each)

**Procedures**

*Identification of the agent*

Real time RT-PCR

- This method is sensitive and specific, and no amplification is observed with any of the related 27 serotypes of BTV
- Commercial real-time RT-PCR kits based on genome segment 9 are available and are widely used. They detect all known serotypes of EHDV, possibly as early as 2 days post-infection
- Serotype-specific real-time RT-PCR based on segment 2 are available to identify each serotype

RT-PCR

- Allows the detection of EHDV RNA in blood samples and other tissues
- Serotype-specific RT-PCRs targeting segment 2 of the viral RNA have been developed, as well as multiplex real-time RT-PCRs for the discrimination between EHDV and BTV
- Although RT-PCR has high sensitivity and specificity, RT-PCR based diagnosis should be interpreted with caution: the RT-PCR technique detects viral RNA with a very high level of sensitivity, but this does not necessarily indicate the presence of infectious virus
- The capacity of RT-PCR assays to detect very small numbers of nucleic acid molecules means that such tests are exquisitely sensitive to contamination by extraneous nucleic acids

Isolation in cell culture

- Virus isolation can be attempted from the blood of viraemic animals, tissue samples including spleen, lung and lymph nodes of infected carcasses, and from *Culicoides* spp.
- EHDV can be isolated by inoculation of cell cultures such as those of cattle pulmonary artery endothelial, BHK-21, and Vero

- Unlike Bluetongue virus, embryonated chicken eggs are less sensitive for EHDV isolation. *Aedes albopictus* and *Culicoides variipennis* cell lines may also be used for virus isolation

*Serological tests*

Competitive ELISA (C-ELISA) (serogroup specific)

- The EHD C-ELISA was developed to measure EHDV-specific antibody without detecting cross-reacting antibody to other orbiviruses. These techniques, making use of MAbs against EHDV VP7, are able to detect EHDV serogroup-specific antibodies, and currently are the preferred technique

Virus neutralisation (VN) (serotype specific)

- The gold standard for the identification and quantification of antibodies against EHDV serotypes present in test samples is the VN test
- Detects and quantifies serotype-specific antibodies
- Its main disadvantage is that all suspected virus serotypes must be included in the assay. Consequently, it can be a very time consuming and labour intensive test to perform
- The VN test requires 4–6 days to be completed

Agar gel immunodiffusion

- In the past this test was used for animal trade. It is simple, economical and the antigen used in the assay is relatively easy to generate
- The disadvantage of the AGID is its lack of specificity in that it cannot discriminate between BTV and EHDV. AGID positive sera should be retested using a serogroup-specific test, at least in those areas where BTV and EHDV may be co-circulating
- The AGID can detect antibodies from 5–15 days after infection to 2 years or more

Complement Fixation Test (CFT) (serogroup specific)

- Sensitive and specific for EHDV diagnosis
- It was used until 1980 for diagnosis and certification of animals for export
- The test is serogroup-specific and inexpensive with a sensitivity similar to the VN and AGID tests
- CFT allows detection and quantification of antibodies for 4–12 months after infection but is less reliable after this period

**For more detailed information regarding laboratory diagnostic methodologies, please refer to Chapter 3.1.7 Epizootic haemorrhagic disease in the latest edition of the *OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals* under the heading "Diagnostic Techniques".**

## *PREVENTION AND CONTROL*

- Other than Ibaraki in cattle, treatment and control is limited for EHDV

### *Sanitary prophylaxis*

- Protect animals during loading and transport operations, both by air and land, through physical barriers, insect repellents and planning of operations for low vector activity times of the day
- Management of *Culicoides* breeding areas near cattle hosting facilities

### *Medical prophylaxis*

- Both live modified and inactivated vaccines have been developed to control Ibaraki disease in cattle in Japan

- In USA, vaccines were developed for captive wildlife deer farmers. These are autogenous inactivated vaccines from EHDV isolates originating from ill or dead animals in affected premises. Their use must be approved by government authorities

**For more detailed information regarding safe international trade in terrestrial animals and their products, please refer to the latest edition of the *Terrestrial Animal Health Code*.**

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The OIE will periodically update the OIE Technical Disease Cards. Please send relevant new references and proposed modifications to the OIE Scientific and Technical Department (scientific.dept@oie.int). Last updated December 2019