ENCEPHALOMYOCARDITIS VIRUS

Prepared for the Swine Health Information Center
By the Center for Food Security and Public Health,
College of Veterinary Medicine,
Iowa State University
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SUMMARY

Etiology

- Encephalomyocarditis virus (EMCV) is a non-enveloped RNA virus in the family *Picornaviridae*.
- Multiple strains are known but most belong to a single serotype, EMCV-1. A second serotype, EMCV-2, was identified in 2012.

Cleaning and Disinfection

- EMCV is resistant to many environmental conditions and remains stable at pH 3–8. The virus is inactivated at low humidity levels (below 50%) and a temperature of 60°C after 30 minutes.
- Water containing 0.5ppm chlorine, iodine based disinfectants, and mercuric chloride have been used as disinfectants for EMCV.

Epidemiology

- EMCV has a broad host range and has been isolated in over 30 species of mammals and birds. Rodents are thought to be the reservoir species.
- EMCV has a worldwide distribution. Seasonal outbreaks have been reported.
- EMCV was recognized as a swine pathogen in 1958. Clinical cases have been reported in domestic swine in Europe, Canada, South America, Australia, Korea, China, and the United States (Hawaii, Illinois, and Iowa).
- In neonatal pigs, mortality rates can reach 100%.
- Infections in humans appear to be mainly asymptomatic. Pig-to-human transmission has not been documented but remains a concern where pigs are used as donors for human xenografts.

Transmission

- Ingestion, either of EMCV-infected carcasses (rats or mice) or of food/water contaminated by infected carcasses, is thought to be the primary route of transmission in swine. Direct pig-to-pig transmission has not been demonstrated.

Infection in Swine/Pathogenesis

- Sudden death can occur in neonates. Lethargy, fever, anorexia, dyspnea, vomiting, and paralysis have also been reported.

- In older animals, infection is usually asymptomatic. Fever and myocarditis can be seen. Abortion is common in gestating sows.

Diagnosis

- EMCV can be isolated from a number of cell lines, chicken embryos, and mouse embryos.
- Reverse transcriptase polymerase chain reaction (RT-PCR) is the most common method of detection; a recently developed reverse transcriptase loop-mediated isothermal amplification method (RT-LAMP) shows promise for use in the field.
- A variety of serological tests are available including virus neutralization and enzyme-linked immunosorbent assay (ELISA), which are most frequently used.

Immunity

- An inactivated vaccine for EMCV is available. Virus-like particle (VLP) vaccines are also being investigated for use in swine.

Prevention and Control

- Prevention of EMCV infection is based on rodent control; for example, methods to reduce attraction of rodents include keeping farms clean, removing trash, and cleaning up feed spills. Rodents can be excluded from buildings by sealing cracks and areas around water pipes and electrical wires. Baiting and trapping can also reduce rodent populations.

Gaps in Preparedness

- Transmission routes for EMCV are poorly understood. The potential for transmission of EMCV from pigs to humans via xenografts should continue to be investigated.

OVERVIEW

Encephalomyocarditis virus (EMCV) is a ubiquitous virus with a broad host range. Commonly considered a rodent pathogen, EMCV is known in the swine industry to cause acute myocarditis with sudden death in young pigs and abortion in gestating animals. The virus was recognized as a swine pathogen in1958, when it was isolated as the causative agent of acute death in a pig in Panama. The first appearance of EMCV in the United States occurred in Florida, during a series of outbreaks from 1960–1966.

EMCV is a non-enveloped, single-stranded RNA virus in the genus *Cardiovirus* of the family *Picornaviridae*. Currently, there is a single recognized serotype, EMCV-1, though a second, EMCV-2, was recently described. Strains are similar in antigenicity and differ only in their hemagglutination activity. Original strains include Columbia-SK virus, MM virus, encephalomyocarditis virus, and Mengo encephalomyelitis virus. Similar to other related picornaviruses, such as foot-and-mouth disease virus (FMDV), EMCV is extremely resistant to environmental conditions, surviving wide temperature and pH ranges. Seasonal outbreaks have been reported, with clinical cases peaking in autumn.

EMCV is unique among picornaviruses in that it has a broad host range and a wide geographic distribution. It has been the causative agent of swine disease outbreaks in Canada, Peru, Belgium, Italy, Republic of Korea, China, and the United States. Despite its reputation as a rodent pathogen, EMCV can infect a variety of mammals, birds, and arthropods. Susceptible domestic animals include dogs, cats, horses, cattle and swine, though most infections are asymptomatic in domestic animals except swine. Several wild and zoo species are susceptible to infection with EMCV; it has been associated with disease outbreaks in elephants, lemurs, non-human primates and llamas.

The virus can be isolated in cell lines originating from primates, rodents, swine, and others. There are several methods available to test for viral antibody, but virus neutralization (VN) and enzyme-linked immunosorbent assay (ELISA) are the most specific and are most commonly used. Experimentally, reverse transcriptase polymerase chain reaction (RT-PCR) has been used successfully, and more recently a reverse transcriptase loop-mediated amplification method has been reported as a rapid, highly sensitive, and specific test that could be implemented in the field. Inactivated commercial vaccines are available for use in domestic and wild animals in the United States.

In swine, rodents are thought to play a role in transmission. Ingestion, either of EMCV-infected carcasses or of food/water contaminated by infected carcasses, is the primary route of transmission. Lethargy, fever, anorexia, and paralysis can be seen with infection, but most often there are no clinical signs prior to sudden death. Stillbirths or mummified fetuses are common with infection in gestating animals. Focal, discrete discoloration of the myocardial tissue is the prominent gross lesion, accompanied by non-suppurative interstitial myocarditis histologically. The duration of infection is short and animals are often found dead before clinical signs are seen.

EMCV is not considered to be a major threat to the United States swine industry; it is not highly contagious, there have been no reported cases of animal-to-human transmission, and a commercial vaccine is available. However, little research has been done regarding transmission of EMCV in pig-to-human xenografts. Experimental studies suggest that xenozoonotic viral infections could result in clinical disease in humans. Further research is needed to establish prophylactic and therapeutic measures in the case of this route of infection.

LITERATURE REVIEW

1. Etiology

1.1 Key Characteristics

Encephalomyocarditis virus (EMCV) is a non-enveloped, single-stranded RNA virus belonging to the genus *Cardiovirus* in the family *Picornaviridae*.[1]

1.2 Strain Variability

Several antigenically similar strains of EMCV were isolated throughout the 1940s. Columbia-SK virus was the first strain discovered in 1940[2], followed by MM virus in 1943[6], encephalomyocarditis virus in 1944[3], and finally Mengo encephalomyelitis virus discovered in 1946[7]. These strains comprise a single serotype, encephalomyocarditis virus-1 (EMCV-1)[1], and cannot be distinguished by cross-neutralization tests, indirect immunofluorescence, complement fixation tests, or Western immunoblotting. Strains do, however, differ in their hemagglutinating activity.[8] In 2012, a second serotype was described, EMCV-2, that can be distinguished from EMCV-1 by serologic and molecular means. [9]

2. Cleaning and Disinfection

2.1 Survival

Similar to other picornaviruses, EMCV isolates are resistant to many environmental conditions. The virus remains stable at a pH range of 3–9.[8] A change of temperature from 10°C to 37°C has little effect on the inactivation of aerosolized EMCV particles. At humidity of 75%, aerosolized EMCV can survive inactivation over a six hour time period, but at a humidity of 50% or less, it is rapidly inactivated.[12]

2.2 Disinfection

EMCV is resistant to ether. It can be inactivated by heating to 60°C for 30 minutes.[10] Water containing 0.5ppm chlorine, iodine based disinfectants, and mercuric chloride can also be used as disinfectants.[13] Alternatively, a femtosecond laser has been reported as a non-invasive and environmentally friendly method of inactivation. [14]

3. Epidemiology

3.1 Species Affected

EMCV was first described in 1940 as a novel rodent-borne virus.[2] In 1944, it was isolated in non-human primates with sudden and unexpected deaths.[3] EMCV wasn't recognized as a swine pathogen until 1958, when the virus was isolated from the spleen and lung of a pig that suffered an acute death in Panama.[4] Shortly after, EMCV was seen in swine in a series of outbreaks in Florida from 1960-1966.[5]

EMCV has a broad host range and has been isolated in over 30 species of mammals and birds.[8] Rats and mice have been proposed as the reservoir species for EMCV; however, experimental studies have been unable to support this claim. Natural transmission between rodents is difficult to achieve and a chronic carrier state has not been observed.[8,15,16] Susceptible mammalian species include, but are not limited to, non-human primates, elephants, squirrels, mongooses, raccoons[15] and lemurs.[11] Domesticated species susceptible to EMCV include dogs, cats, cattle, horses and swine. With the exception of swine, most EMCV infections in domestic animals are asymptomatic. EMCV has also been isolated from several species of arthropods, although experiments have not demonstrated that they play a role in transmission.[8]

3.2 Zoonotic Potential

Neutralizing antibodies have been found in humans throughout the world, indicating infection is common. However, clinical disease is rare, suggesting frequent undiagnosed or asymptomatic disease.[16] In 2009,

two human cases were confirmed in Peru. The patients presented with fever, inappetence, and headaches.[17] There have been no reports of animal-to-human transmission of EMCV.[8] If pigs are to be used as donors for human xenografts in the future, transmission of this virus from EMCV infected pigs to humans, should not be ruled out.[10]

3.3 Geographic Distribution

EMCV has a worldwide distribution. Clinical cases have been reported in domestic swine in Europe[18], Canada[19-21], South America[22], Australia[23], Korea[24], China[25], and the United States.[5,26,27] Within the United States, seropositive pigs have been found in Hawaii[15], Illinois[28], and Iowa[29,30].

Seasonal outbreaks of EMCV have been recorded, with the peak occurring during cooler months.[10,11]

3.4 Morbidity and Mortality

Clinical disease due to infection with EMCV is uncommon in most domestic animals, with the exception of swine. Mortality is seen in animals ranging from 4 days to 24 weeks, with higher mortality rates in younger animals.[23,31-33] In pigs less than one week of age, mortality rates of 100% have been reported.[34] Specific data on morbidity in swine due to EMCV is unavailable.

4. Transmission

Under experimental conditions, many routes of exposure (intranasal, intramuscular, intratracheal, oral, aerosol, subcutaneous, intracranial) have resulted in transmission of EMCV in susceptible species.[8] Zimmerman et al.[35] demonstrated that transmission via contaminated wounds in swine is possible, but direct pig-to-pig transmission was not achieved. A case-control study found that swine farms with large populations of mice were more likely to have clinical EMCV infections than farms with few or no mice, indicating rodents play a role in transmission.[11,33] Natural transmission of EMCV in swine is likely due to ingestion of carcasses infected with EMCV (usually mice or rats), or ingestion of food or water contaminated by EMCV-positive carcasses.[15]

5. Infection in Swine/Pathogenesis

After oral introduction, EMCV travels to the tonsils where it is introduced to monocytes. EMCV then spreads to target organs via monocytes. The main target organ for EMCV in swine is the heart. It has also been found in endothelial tissue of virtually every organ, including spleen, kidney, intestine, pancreas, lung, and lymph nodes.[31,36]

5.1 Clinical Signs

Severity of clinical signs are dependent upon several factors including environment, co-existing infections, drugs/chemicals, and host and viral characteristics.[33,36] In younger, more susceptible animals, sudden death often occurs without any accompanying clinical signs. Death occurs between 2–11 days post-infection (more commonly 2–4 days).[8,10,32] Lethargy, fever, anorexia, dyspnea, vomiting, and/or paralysis have also been reported.[20,23,32] Older, less susceptible animals may be asymptomatic or exhibit mild illness with a fever and myocarditis. Abortion is common in gestating animals with EMCV infection. [20,31,33,36]

5.2 Postmortem Lesions

In swine, the most common gross lesion associated with acute death, is focal, white discoloration of the myocardium. It may be accompanied by heart dilation, hydropericardium, pulmonary edema, ascites, and/or hydrothorax.[31] Focal non-suppurative interstitial myocarditis, with mononuclear cellular infiltrates, can be seen histologically.[23,31,36,37] Necrotizing tonsillitis and focal interstitial pancreatitis are additional histologic lesions that have been seen in some cases.[31]

6. Diagnosis

6.1 Clinical History
Sudden death in younger animals and abortion in gestating animals are suggestive of EMCV.[8]

6.2 Tests to Detect Nucleic Acids, Virus, or Antigens
Virus isolation is most commonly done using baby hamster kidney fibroblasts (BHK-21), African green monkey kidney (Vero), and human cancer (HeLa) cell lines, but EMCV also replicates well in mouse and chicken embryos. A fluorescent antibody test, using an anti-EMCV fluorescently-conjugated antibody, is available for virus identification.[10]

RT-PCR has been reported as a molecular method of detection.[38] More recently, reverse transcriptase loop-mediated isothermal amplification method (RT-LAMP) has been reported as a rapid, specific, and sensitive test that can be easily utilized in the field.[39]

6.3 Tests to Detect Antibody
Serum antibody can be detected using hemagglutination inhibition (HI), virus neutralization (VN), enzyme-linked immunosorbent assay (ELISA), immunofluorescent antibody assay (IFA), and agar-gel immunodiffusion (AGID). VN and ELISA are used most frequently and have shown to be the most specific, detecting antibody titers greater than or equal to 1:16.[10]

6.4 Samples
6.4.1 Preferred Samples
Preferred samples for EMCV isolation are the heart and brain.[40]

6.4.2 Oral Fluids
Specific data regarding the presence of EMCV in oral fluids is unavailable.

7. Immunity

7.1 Post-exposure
Neutralizing antibody can be detected in serum as early as 2–3 days post-infection[41] and can persist for six months to a year. Maternal antibody is protective until approximately two months of age.[10]

7.2 Vaccines
Inactivated EMCV vaccines are available for use in domestic and wild animals in the United States.[10] Virus-like particle vaccines, produced with the baculovirus expression system, have recently been studied. Results indicate VLP-vaccines are a safe option that would provide high antigenicity and immunogenicity in swine.[42]

7.3 Cross-protection
Currently EMCV has only one recognized serotype, EMCV-1, although a second serotypes, EMCV-2, has recently been described.[9] Strains of EMCV-1 have little antigenic variation and therefore, cross-protection between strains is likely.[10]

8. Prevention and Control
There is no treatment for EMCV infection. In endemic areas, rodents are thought to play a role in transmission and accordingly, prevention and control measures should focus on keeping rodent populations to a minimum.[8,10] Movement of manure through slatted floors and movement between pits

were shown to be significantly protective in swine populations exposed to EMCV.[33] To protect against clinical disease, a commercially available, inactivated vaccine can be administered to susceptible animals.

9. World Organization for Animal Health (OIE) Terrestrial Animal Health Code

EMCV is not included in the 2015 OIE Terrestrial Animal Health Code. There are no restrictions for importation of animals from countries or zones infected with EMCV.

10. Gaps in Preparedness

EMCV is not considered to be a highly contagious pathogen, although the routes of transmission are not completely understood. Rodents are involved in transmission of EMCV but the full extent of their role is unclear. Direct transmission of the virus has been confirmed in some experimental studies[7] and refuted in others.[32] A clearer understanding of the transmission of this pathogen among swine populations will benefit prevention and control efforts.

Clinical disease from EMCV infection in humans is rare, but the potential use of pigs for human xenografts calls for more research in this area. Brewer et al.[43] showed that intra-abdominal transplantation of pig myocardial sections, acutely infected with EMCV, into mice resulted in infection and acute fatal disease. This reveals the need for development of prophylactic and therapeutic measures for possible pig-to-human viral xenozoonotic infections.

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Authors, Center for Food Security and Public Health, College of Veterinary Medicine, Iowa State University:

- Sarah Horak, BS; 2nd year student,
- Kristin Killoran, PhD; 2nd year student
- Kerry Leedom Larson, DVM, MPH, PhD; Veterinary Specialist

Reviewers, Center for Food Security and Public Health, College of Veterinary Medicine, Iowa State University:

- Pamela Zaabel, DVM; Veterinary Specialist
- James A. Roth, DVM, PhD; Director

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