MODULE-21: RETROVIRUS - EQUINE INFECTIOUS ANAEMIA

Learning objectives

- Group VI Reverse transcribing RNA viruses
  - About the nature and morphology of EIAV
  - Pathogenesis of EIAV
  - About the diagnosis and various lab tests
  - About vaccines
  - About the various tests and regulation for international trade
  - About the control and eradication of EIAV

EQUINE INFECTIOUS ANAEMIA
INTRODUCTION

- Equine infectious anemia (EIA) is an infectious and potentially fatal viral disease of members of the horse family. EIA is significant historically because it is the first disease of horses proven to be caused by a “filterable virus”. EIAV is the first lentivirus–induced disease proven to be transmitted by insects. And EIA is the first persistent virus for which antigenic drift was defined. EIA is also the first lentivirus–induced disease for which a diagnostic test was approved.

EQUINE INFECTIOUS ANAEMIA
GENERAL ASPECTS

- Explains classification, morphology, replication etc.

EQUINE INFECTIOUS ANAEMIA
CLASSIFICATION

Baltimore group	Group VI Reverse transcribing RNA viruses
Family	Retroviridae
Sub-family	Orthoretrovirinae
Genus	Lentivirus

Species	Equine infectious anaemia virus (EIAV)

EQUINE INFECTIOUS ANAEMIA
SYNONYMS

- Swamp fever.

EQUINE INFECTIOUS ANAEMIA
MORPHOLOGY

- The virions have a complex construction and consist of an envelope, a nucleocapsid, a nucleoid, and a matrix protein. Virions are enveloped, spherical to pleomorphic in shape and measure 80-100 nm in diameter. The surface has numerous glycoprotein spikes, which are 8nm in length. The core is rod-shaped and the nucleoid is concentric. The genome is dimeric (diploid) comprises of two monomers. The genome consists of non-segmented two molecules of linear positive-sense, single-stranded RNA joined by hydrogen bonds. Minor species of non-genomic nucleic acid are also found in virions. The genome is composed of 9200 nucleotides. The 5'-end of the genome has a methylated cap and the 3'-terminus has a poly (A) tract. The genome of retroviruses is unique in the following aspects.
    - They are the only viruses, which are truly diploid.
    - They are the only RNA viruses whose genome is produced by cellular transcriptional machinery
    - They are the only viruses whose genome requires a specific cellular RNA (tRNA) for replication.
    - They are the only (+) sense RNA viruses whose genome does not serve directly as mRNA immediately after infection. Hence, the nucleic acid is not infectious
- The genome is comprised of four genes gag, pro, pol and env. Each gene codes for a single or more than one protein. The important proteins are listed below.

Name	Protein	Function
MA	Matrix matrix protein (gag gene)	lines envelope
CA	Capsid capsid protein (gag gene)	• protects the core • most abundant protein in virus particle
NC	Nucleocapsid capsid protein (gag gene)	protects the genome; forms the core
PR	Protease	Essential for gag protein cleavage during maturation
RT	Reverse transcriptase (RNA dependant DNA polymerase)	Reverse transcribes the RNA genome
IN	Integrase Encoded by the pol gene	needed for integration of the provirus
SU	Surface glycoprotein	The outer envelope glycoprotein;

		major virus antigen
TM	Transmembrane protein	The inner component of the mature envelope glycoprotein

EQUINE INFECTIOUS ANAEMIA
OIE LISTING AND RISK GROUP

- *OIE Classification*: List B infection
- *Risk group*: No known human infection.

EQUINE INFECTIOUS ANAEMIA
RESISTANCE

- EIAV are fragile. They are sensitive to treatment with heat, detergents, and formaldehyde. Their infectivity is not affected by irradiation. The virus lasts for several months at room temperature, in urine, faeces, and dried blood etc.

EQUINE INFECTIOUS ANAEMIA
REPLICATION

rs inside the cytoplasm and it is the only RNA virus that depends on host cell transcriptionary mechanisms. The viru
a host cell receptor. The SU and TM proteins help in this stage. The virus enters by fusion. Only partial uncoating oc
gh the RNA is positive sense they are not translated immediately inside the cytoplasm.
A is synthesized inside the cell with a virus coded enzyme RNA dependant RNA polymerase (reverse transcriptase).
eaction is known as the provirus. Three forms of provirus DNA are found in all infected cells. This DNA later migrate
ost cell DNA with the help of the enzyme integrase. Retroviruses use the cellular transcriptional machinery for expre
e cellular genes.
s at the cytoplasm and the mature virions later bud out through the cell membrane, acquiring the envelope during th
ion steps, the replication processes of retroviruses are error prone. Further changes in the retro viral genome occurs
bination and exchange of genes with host genome.

EQUINE INFECTIOUS ANAEMIA
HA PROPERTY

- The virus has no HA property.

EQUINE INFECTIOUS ANAEMIA
STRAINS AND SEROTYPES

- No distinct serotypes are present.

EQUINE INFECTIOUS ANAEMIA
CULTIVATION

- The virus is cultivated in susceptible horses or in leukocyte cultures. The presence of virus in theses systems is confirmed by antigen confirmation tests.

EQUINE INFECTIOUS ANAEMIA
PATHOGENESIS

- Describes host affected, transmission, symptoms and lesions

EQUINE INFECTIOUS ANAEMIA
HOSTS AFFECTED

- The infection is limited to members of the family equidae. Donkeys are less severely affected than horses.

EQUINE INFECTIOUS ANAEMIA
DISTRIBUTION

- The infection is worldwide. Few countries like Australia and New Zealand have claimed freedom.

EQUINE INFECTIOUS ANAEMIA
TRANSMISSION

- EIA is considered a classic bloodborne infection. The infection occurs in summer and autumn since the insects appear in large numbers in these seasons. The spread of EIA is usually slow although it can be quite fast where large groups of horses are gathered in a small area. EIA is very easily spread by the injection of small quantities of infected blood.
- The EIAV most frequently is transmitted between horses in close proximity by large biting insects, such as horseflies and deerflies. The bites from these flies stimulate defensive movement by the horse, which often results in an interruption of the blood–feeding. When interrupted, the fly is motivated to complete the feeding as soon as possible. It then attacks the same or a second host and feeds to repletion.
- In this manner, any infective material from the blood of the first host that is present on the mouthparts of the insect can be mechanically transmitted to the second host. The virus can survive only less than 4 hours in the insect.
- Insect transmission of EIAV is dependent on the number and habits of the insects, the density of the horse population, the number of times the insect bites the same and other horses, the amount of blood transferred between horses, and the level of virus in the blood of the infected horse from which the initial blood meal was obtained.
- Iatraogenic transmission is also possible during any manipulation that may involve the transfer of infected blood or saliva, for example through the use of unsterilised instruments or equipment. Vertical transmission (in utero or through the milk) is also possible

EQUINE INFECTIOUS ANAEMIA
SYMPTOMS

- The first attack is usually the most acute with the horse suffering from depression, profound weakness, and loss of condition. Intermittent fever with large and rapid swings in temperature may also occur. Jaundice and bleeding around the mouth and nasal areas may also occur. This form of the disease is the most damaging and the most difficult to diagnose because the signs appear rapidly, and often only an elevated body temperature is noted. One–fifth of a teaspoon of blood from a horse with acute EIA contains enough virus to infect 1 million horses.
- Animals may show a temporary recovery and appear normal for 2 - 3 weeks and then relapse with similar, but less sever symptoms. It is also referred as chronic form. The symptoms include fever, petechial hemorrhages on the mucous membranes, depression, weight loss, dependent edema under the skin in the legs and under the chest and other underbody surfaces and anemia. The animal may also have an irregular heartbeat, and a jugular pulse. One fifth of a tea spoon of blood from a chronic case during a feverish episode contains enough virus to infect 10,000 horses.
- Most horses also remain as inapparent carriers. These horses show no obvious clinical signsas a result of infection. They survive as reservoirs of the infection for extended periods. Inapparent carriers have dramatically lower concentrations of EIAV in their blood than horses with active clinical signs of the disease. Only 1 horsefly out of 6 million is likely to pick up and transmit EIAV from this horse. All horses infected with EIAV are thought to remain virus carriers for life. The inapparent form may become chronic or acute due to severe stress, hard work, or the presence of other diseases.

EQUINE INFECTIOUS ANAEMIA
LESIONS

- In horses dying of the disease there is haemorrhages in the liver, spleen, kidneys, on serous membranes and in the mucosa of the intestines. The liver is enlarged and often yellowish-brown in colour. The spleen is also enlarged with soft pulp.The heart muscle appear pale, flabby and friable

EQUINE INFECTIOUS ANAEMIA
DIAGNOSIS

- Dealt as Field and Laboratory diagnosis

EQUINE INFECTIOUS ANAEMIA
FIELD DIAGNOSIS

- Field diagnosis is based on the Coggins test. Since, no vaccines are available against EIAV, presence of antibodies against EIA in the serum of horses is an indication of infection. Clinical signs and lesions also provide clue about EIA.

EQUINE INFECTIOUS ANAEMIA
LABORATORY DIAGNOSIS

- *Isolation and identification*: Rarely attempted since it is of no practical value. Blood samples should be collected to confirm the diagnosis using the Coggins (agar gel diffusion) test or the more recent ELISA test. Isolation of the virus from suspect horses

may be made by inoculating their blood on to leukocyte cultures prepared from horses free of infection. Virus production in cultures can be confirmed by detection of specific EIA antigen by ELISA, or imunofluorescence assay.

- *Nucleic acid identification method*: Recently a RT-PCR method has been developed to detect EIA proviral DNA from the peripheral blood of horses

EQUINE INFECTIOUS ANAEMIA
SEROLOGY

- Due to the persistence of EIA virus in infected equids, detection of serum antibody to EIA virus confirms the diagnosis of EIA virus infection.
    - *Coggins test – (Agar gel immunodiffusion test):* It is a test prescribed for International trade. Precipitating antibody is rapidly produced as a result of EIA infection, and can be detected by the AGID test. Specific reactions are indicated by precipitin lines between the EIA antigen and the test serum and confirmed by their identity with the reaction between the antigen and the positive standard serum.
    - *ELISA*: Competitive and non-competitive ELISAs are also used to identify antibodies against EIAV.

EQUINE INFECTIOUS ANAEMIA
DIFFERENTIAL DIAGNOSIS

- EIA should be differentiated from purpura haemorrhagica, babesiosis, leptospirosis, severe strongyliasis or fascioliasis, phenothiazine toxicity and auto-immune haemolytic anaemia

EQUINE INFECTIOUS ANAEMIA
TREATMENT

- There is no effective treatment available against EIA. Supportive treatment, including blood transfusions can be considered. However, because recovered horses can become carriers, if the disease occurs in a free area it is generally better to destroy affected horses.

EQUINE INFECTIOUS ANAEMIA
CONTROL AND ERADICATION

- Vaccines
    - There are no vaccines against EIA
- Eradication
    - Eradication is aimed in minimizing or eliminating contact of horses with the secretions, excretions, and blood of EIAV–infected horses. This has to be carried out by testing and segregating test–positive horses from those with negative test results. Once the reservoirs of EIAV are identified, separated, and maintained a safe distance from the other horses.
    - As recovered horses continue to carry the virus, the most likely source of introduction of EIA is through importation of horses. OIE guidelines suggest

that, when importing horses, as well as showing no clinical signs, they should be tested negative during the 30 days prior to shipment.
    - In endemic areas, risk of infection can be reduced by protecting horses from insect vectors by
        - Keeping horses away from low lying areas
        - Providing draining swamps
        - Through use of insecticides and by providing insect-proof stabling

EQUINE INFECTIOUS ANAEMIA
ADDITIONAL READING

- By following precautions, horse owners can get rid of the infection. These precautions are specified by the USDA, APHIS, Veterinary Services, National Center for Animal Health Programs, Certification and Control Team
    - Use disposable syringes and needles. Follow the rule of one horse—one needle.
    - Clean and sterilize all instruments thoroughly after each use.
    - Keep stables and immediate facilities clean and sanitary. Remove manure and debris promptly, and ensure that the area is well drained.
    - Implement insect controls. The local veterinarian or animal health official can provide information about approved insecticides and other insect–control measures. Avoid habitats favorable to insect survival.
    - Do not intermingle infected and healthy animals. Do not breed EIAV–positive horses.
    - Isolate all new horses, mules, and asses brought to the premises until they have been tested for EIA.
    - Obtain the required certification of negative EIA test status for horse shows, county fairs, race tracks, and other places where many animals are brought together.
    - Abide by State laws that govern EIA.