

# **Equine Influenza Virus (EIV) Disease Guidelines**

#### **Definition**

Equine influenza virus (EIV) is an RNA virus endemic to equine populations in many countries worldwide. Equine influenza outbreaks may also occur sporadically in epidemic form. Countries historically free of equine influenza include Iceland and New Zealand. While epidemic outbreaks have occurred in Australia, Japan, and South Africa, these countries are currently considered equine influenza-free.

Equine influenza virus has historically existed in 2 forms: the H7N7 or equine-1 subtype, and the H3N8 or equine-2 subtype. The equine-1 subtype is believed to be extinct and is no longer recommended for use in vaccines. The equine-2 subtype undergoes a gradual process of change (antigenic drift) due to accumulating mutations in its major surface antigens, the hemagglutinin (H or HA) and neuraminidase (N or NA). This drift sometimes leads to the co-existence of virus strains belonging to lineages that once were very similar but have drifted to become increasingly dissimilar, different enough to make cross-immunity ineffective. That is, a given horse's level of immunity to equine influenza might be sufficient to protect against one lineage but not the other. Because horses travel internationally, this necessitates the inclusion of representative strains from both viral lineages in equine influenza vaccines. Currently there are 2 such co-circulating lineages, called Florida clade 1 and Florida clade 2, that split around the year 2000 and have been drifting further apart ever since. A vaccine against one lineage does not guarantee protection against the other, and thus both are recommended for inclusion in vaccines. For many years the Florida clade 1 lineage dominated the EIV circulation in the USA and the Florida clade 2 lineage dominated in Europe. Since 2018–19, the Florida clade 1 has been dominant in Europe as well, while the Florida clade 2 lineage has persisted most notably in China.

# **Incubation Period**

The period between exposure to EIV and appearance of clinical signs is frequently as short as 24 hours but may be up to 3 days. Asymptomatic infections in partially immune horses may produce a lag period (one or more weeks) between arrival of the virus source and the observed start of an outbreak.

#### **Clinical Signs**

Clinical signs vary in their severity depending on the age and immune status of the horse, and asymptomatic infection is possible. Clinical signs are more common and often more severe in younger horses (ages 1–5yo). Older horses generally have milder disease. Equine influenza may be more severe in donkeys and mules.

- Fever, up to 106F (41.1C)
- Lethargy
- General malaise
- Anorexia
- Muscle pain/weakness

- Dry, harsh cough (sometimes paroxysmal) that usually precedes fever. The dry, harsh cough is a frequent clinical sign in EI, however, not all horses with EI will develop a cough.
   Cough can last up to 6 weeks after all other clinical signs have abated and affected horses may take up to 6 months to regain previous athletic abilities.
- Mild lymphadenopathy of the retropharyngeal and submandibular lymph nodes.
- Serous nasal discharge that may progress to mucopurulent as a result of secondary bacterial infection.
- Secondary bacterial infections are common in influenza-affected horses due to mucociliary apparatus destruction. If left untreated, these can cause life-threatening pleuropneumonia.
- Rarely, clinical signs may include distal limb edema and signs consistent with cardiomyopathy.

#### **Risk Factors**

- Age: horses 1–5 years of age, although all ages are at risk
- Areas of high co-mingling of horses such as racetracks, show grounds, veterinary hospitals
- Immunosuppression from traveling, hospitalization, training, and showing

*Note*: While vaccination reduces the risk of clinical disease, vaccinated horses can still become infected and shed virus (subclinical shedding)

### **Transmission and Shedding**

Respiratory transmission occurs most commonly through inhalation of infective droplets from coughing and sneezing horses. The distance these droplets may spread through the air has not been definitively established but may be as far as 50 yards (approximately 45 meters).

Indirect transmission can occur and is likely an important means of spread. This includes transmission of the virus on contaminated clothing, equipment, brushes, shared water buckets, hands, etc.

Respiratory shedding of contagious virus typically lasts for 7–10 days post-infection in naïve animals; much shorter shedding periods (e.g., 5 days) occur in partially immune (previously vaccinated) horses. Horses may be PCR-positive on nasal secretions beyond the period for which they are infectious, often 2 weeks or more.

# **Diagnostic Sampling, Testing and Handling**

### Virus isolation from nasopharyngeal swabs

- Samples should be collected within 24–48 hours after onset of clinical signs.
- Swabs should be submitted in viral isolation transport media (NOT bacterial transport media). If no viral transport media is available, place swabs in a red top tube with at least 1 cc of sterile saline (enough to keep the tip of the swab immersed). Samples should be shipped overnight on ice.



• Nasopharyngeal swabs are a superior alternative to nasal swabs, in that these collect about 10 times as much virus as nasal swabs which reduces the possibility of a false-negative diagnostic test result. These are also safer in that there is no chance that horse movement plus inhalation might cause the swab to be lodged inaccessibly within the nasal meatus. However, the short (Q-Tip) type swab is preferable to no swab at all.

# Real-time PCR (RT-PCR) from nasopharyngeal swabs

- Different diagnostic laboratories may test for different targets, e.g., NP or M targets for pan-influenza A, or H3 HA specific for EIV). Practitioners with a case of suspected influenza that tests negative for H3 HA may want to consult with their diagnostic laboratory about additional influenza testing for other strains.
- EDTA blood is NOT an acceptable sample for diagnosis of acute infection as EIV-infected horses do not have detectable viremia.

# Serology

- Paired sera can be very useful in confirming a diagnosis of equine influenza. The acute sample should be obtained as close to onset of clinical signs (max of 3 days) as possible and convalescent sample should be collected 2 weeks later.
- Serology can be used to confirm infection even in the face of a false negative virus isolation.
- Submit separated serum samples (clot must be removed) in a red top tube. Serum samples
  are stable at room temp for several days; longer requires refrigeration or freezing. The
  acute serum sample can be frozen until the convalescent sample is collected, then both
  shipped together.

# Immunoassay (stall-side kit)

• There are several available products with varying diagnostic reliability. These also require nasopharyngeal swab samples.

#### **Postmortem**

In the USA, it is very rare that uncomplicated EIV infection would result in a fatal outcome. Thus, there are few reports of gross pathologic findings. Based on original studies of influenza, changes include bronchiolitis, peribronchiolitis, and subacute interstitial pneumonia. Practitioners performing necropsies in the field are encouraged to contact a veterinary diagnostic laboratory to which they plan to submit samples for further testing, such as histopathology and pathogen identification in order to be certain they collect the appropriate samples and handle the samples in a manner that will optimize making a definitive diagnosis.

### **Environmental Persistence**

- EIV can remain viable for up to 2 days on contaminated fomites and solid environmental surfaces, e.g., grooming supplies, stall latches, etc.
- EIV can survive in aerosols for several hours and on hands for a few minutes.
- In water, EIV viability has been reported up to 3 days, however, the virus survival in water is temperature dependent and may be longer in cold water.



# **Specific Control Measures**

# Vaccination

- AAEP Equine Influenza (EIV) Vaccination Guidelines
- While annual vaccination is currently recommended, more frequent vaccination may be recommended for young horses and horses at increased risk due to environmental and/or management factors.
- In an outbreak situation, booster vaccination of unexposed, previously vaccinated, healthy animals is unlikely to be of value unless it can be administered at least 10 days prior to exposure.
- If animals are unvaccinated prior to an outbreak, the use of a modified live intranasal vaccine may be recommended to achieve partial protection within 5 days of primary administration.

# **Isolation and Biosecurity**

- The <u>AAEP Biosecurity Guidelines</u> have detailed recommendations on how to prepare for and respond to an infectious disease outbreak.
- Horses showing clinical signs of any respiratory disease (coughing, nasal discharge, fever) should be immediately isolated and standard respiratory biosecurity guidelines should be followed until a diagnosis is confirmed.
- During an influenza outbreak, affected and exposed animals should be isolated from susceptible horses for 14 days, preferably in a different air space. Coughing horses can aerosolize the virus and transmit infective virus particles. The distance these droplets may spread through the air has not been definitively established but may be as far as 50 yards (approximately 45 meters), depending on housing conditions and ventilation.

#### Release of Animals from Isolation

Maintain quarantine and isolation procedures (primary perimeter) for 14 days <u>after</u> resolution of the last suspected case.

### **Biosecurity Issues for Receiving Animals**

Isolate all horses returning from shows, exhibitions, or trail rides for at least 14 days, monitoring temperature at least once per day. Some facilities may want to consider a vaccination requirement if they are at an increased risk for exposure and/or disease.

#### Disinfection

EIV is easily killed by many commonly used disinfectants. Virkon™ S with potassium peroxymonosulfate and sodium chloride kills EIV in most situations and alcohol-based hand sanitizers are effective against influenza viruses.

#### **Zoonotic Potential**

None known, but equine H3N8 influenza virus can infect canines. While equine influenza has not been shown to cause disease in humans, serological evidence of infection has been described primarily in individuals with an occupational exposure to the virus. There is little risk to public

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health. In experimental settings, the virus has shown the ability to infect humans, and a few people in contact with infected horses developed antibodies to equine influenza viruses, but no humans naturally exposed to the virus have become ill, so far as is known.

Some horses have been found seropositive for Influenza D virus, more commonly found in swine and bovines. To date there is no evidence that Influenza D virus causes acute disease in horses. Bovines, cats, humans, and other species have now been shown to also be susceptible to H5N1 avian influenza virus. There have been no known equine cases as of yet (February 2025) and the risk to horses is unknown.

### **Further Reading**

Equine Influenza Info – World Organisation for Animal Health (WOAH) <a href="https://www.woah.org/en/disease/equine-influenza-2">https://www.woah.org/en/disease/equine-influenza-2</a>

Paillot, R. et al. <u>Duration of equine influenza virus shedding and infectivity in immunised horses after experimental infection with EIV A/eq2/Richmond/1/07</u>. Vet Microbiol 166, 22-34 (2013).

Chambers, T.M. and Reedy, S.E. Ch.32: *Equine Influenza Diagnosis: Sample Collection and Transport*. In. E. Spackman (ed.) Animal Influenza Viruses. Methods in Molecular Biology v.1161, pp. 371-377, Springer, New York, 2014.

Reviewed and revised by: Thomas M. Chambers, PhD

**Supported and reviewed by:** AAEP Infectious Disease Committee