AAEP Guidelines: Suspected Case of Clostridial Diarrhea

**Suspected Case of Clostridial Diarrhea**

- **Establish Biosecurity Perimeter**
  - Identify Primary Perimeter
  - Implement Primary Perimeter
    - Stop horse movement
    - Limit human movement
  - Biosecurity Guidelines

- **Communicate the plan**
  - Event management
    - Horsemen
    - Veterinarians
    - Regulatory Agencies
    - Media
    - Related industry members

- ** Attempt diagnosis**
  - Tests
    - Laboratory selection
    - Testing supplies
    - Sample collection
    - Sample handling
    - Sample transport

**Diagnosis**
- Proceed to disease-specific guidelines
  - Salmonellosis
  - Clostridiosis
  - Potomac Horse Fever
  - Coronavirus

**No Diagnosis**
- Maintain Biosecurity (21-28 days after last case)
  - Expand differential diagnoses
  - Consult infectious disease expert

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### Differential Diagnoses

Primary differential diagnoses of possible infectious agent(s) involved with herd health implications:

- **Salmonella spp.**
- **Clostridial enteritis** – *Clostridium difficile, Clostridium perfringens*
- **Potomac Horse Fever** – *Neorickettsia risticii*
- Rotavirus (RTA)
- **Lawsonia intracellularis**
- Coronavirus (ECoV)

(Nota: Until proven otherwise, respond based on ‘worst case’ scenario: *Salmonella.*)

### Establish Biosecurity Perimeter

- The primary biosecurity perimeter is centered on the location of the contagious disease case(s) and exposed animals. It extends until a barrier to further spread of infection is identified.
- The primary perimeter may encompass the entire equine facility (farm, showground or racetrack), or if site design permits, the perimeter may only contain part of the equine facility (barn/paddock). The perimeter should be clearly defined by physical barriers. Signs should be used to identify the perimeter and control access.

**Note:** More than one primary perimeter may be established if case development warrants and facility design permits.

- The primary perimeter contains all suspected infected animals and animals in immediate contact with them.
- All animals within the primary perimeter should be considered infected and contagious until the outbreak is declared over. Animals are prohibited from exiting the primary perimeter, and biosecurity measures are to be implemented to prevent infectious agents leaving the area.
- If the equine facility is an appropriately designed and managed isolation facility then the primary perimeter will be around this facility.
- If the affected horse was moved from its barn to an isolation facility, a primary biosecurity perimeter should remain around the barn from which the affected horse originated.

### Implement Primary Perimeter

- **Stop horse movement**
  - Affected horses should be moved to a separate isolation facility or confined to their stalls.
Clinically unaffected horses are confined within the primary perimeter and managed in order to minimize the spread of an infectious agent.

- **Disease surveillance**
  - Record rectal temperatures twice daily
  - Physical inspections for clinical signs

- **Limit human movement**
  - Access is limited to essential personnel only—veterinarians/technicians/caretakers
  - All personnel follow biosecurity protocols
  - Security personnel may be employed at perimeter access points

### Biosecurity Guidelines

- If the primary perimeter does not encompass the entire facility, it is appropriate to establish a secondary perimeter which does. All animals within the secondary perimeter are considered free of infection, but are at increased risk of exposure, making enhanced disease surveillance and contagion control measures necessary.

- Should clinical disease become apparent in an animal within the secondary perimeter, reassessment of the extent of the primary perimeter is necessary. The affected animal should be situated within the primary perimeter for continued care.

- Animals should travel into and out of the secondary perimeter only from outside the equine facility and not the primary perimeter. All movement of animals should be under the control of the veterinarian in charge.

- **Increase disease surveillance**
  - Monitor and record rectal temperatures of all horses twice daily
  - Physical inspection for clinical signs

  **Note:** It may be advisable to have these tasks performed by individuals designated by the official veterinarian or event management as opposed to representatives of individual horsemen.

- **Regulate horse movement**
  - Record arrival/departure information including:
    - Date
    - Origination/Destination
    - Carrier information
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Communication

The necessity for timely, coherent and accurate information dissemination to affected parties cannot be overstated.

I. Event Management

• Physical plant modification instructions
  o Barriers: designation of isolation facility, the establishment of the physical perimeter (primary and secondary), and the informing of affected parties as to the location/extent of these is a veterinary and/or regulatory responsibility
  o Disinfection instructions
    ▪ During outbreak
    ▪ Before restocking facility with healthy horses
  o Waste removal: maintain appropriate general hygiene of the facility while ensuring potentially infectious material is secured and handled correctly
  o Vermin control: depending on the infectious agent, fomite spread can compromise perimeter integrity and prolong the outbreak

• Personnel management
  o Requirements: adherence to biosecurity protocols and discretion with information should be emphasized
  o Instructions: all relevant protocols should be distributed to involved personnel. Relevant notifications should be prominently displayed within the designated perimeters
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- Notification of zoonotic risk, if pertinent

**Note:** All cases of equine diarrhea should be considered zoonotic until Salmonella or Clostridium sp. have been ruled out.

_A Review of Equine Zoonotic Diseases: Risks in Veterinary Medicine_ (J.S. Weese)

- Outbreak updates: all affected parties should receive regular updates as to the status of the outbreak. Maintaining confidentiality of information by recipients should be stressed to avoid inaccurate portrayal of the situation to peripheral parties
- Event Management Biosecurity Resources
  - Biosecurity Tool Kit for Equine Events

II. Veterinarians
- Instructions: disease surveillance/testing/reporting is the responsibility of the veterinarian and their designee
- Biosecurity guidelines
- Health requirements: entrance into/exit out of facility will be under veterinary control and supervision
- Outbreak updates: the veterinarian responsible or their designee will be the single point source for updated information to be disseminated to interested parties to ensure a coherent message

III. Horsemen
- Disease information for horsemen/owners: all information to lay parties should be in a language that is easily understandable, relates scientific to lay terminology, and avoids misinterpretation.
- Biosecurity guidelines
- Human exposure/zoonotic risk management: any potential for human infection should be stated in dispassionate language.
- Instructions for caretakers
  - Instructions for caretakers
  - Notification of zoonotic risk, if present
  - Instructions for reporting human disease
- Outbreak updates: should be consistent with information disseminated to veterinary stakeholders.
- Requirements for equine entrance into/exit out of facility: must be clearly stated and equally enforced for all parties.

IV. Regulatory Agencies
- Disease notification
Veterinarians are required to be aware of currently reportable or notifiable diseases to the state veterinarian and abide by state regulations.

Note: State and USDA veterinarians can be useful resources during outbreaks of non-reportable infectious disease.

- Outbreak updates: regular communication with State officials and USDA veterinarians is important where notifiable disease is confirmed

V. Media
- Dissemination of information to horsemen and related industry members:
  - Outbreak updates: coherent and consistent with information to affected parties
  - Requirements for equine import into/export out of facility: close liaison with regulatory officials is essential where animal movement is possible

VI. Related Industries
- Outbreak updates
- Summary of biosecurity measures
- Requirements for equine import into/export out of facility

Complete physical exam with special emphasis on the gastrointestinal system

Attempt Diagnosis

Diagnostic Sampling, Testing, and Handling

Feces
- Primary Testing
  - Targeted or panel screening for enteric pathogens by RT-PCR or qPCR
  - Salmonella-specific culture—indicate on laboratory submission form
- Secondary Testing
  - Aerobic culture and sensitivity
  - Anaerobic culture (including Clostridial culture and toxin testing)
  - ELISA testing for agents or toxins (Rotavirus, Clostridial toxins)
  - Antibacterial susceptibility testing and serotyping of Salmonella spp. Isolate

Whole Blood
EDTA tube: PCR for Potomac Horse Fever
<table>
<thead>
<tr>
<th><strong>Laboratory Selection</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Serology</strong> serum may be used for testing in Potomac Horse Fever and <em>Lawsonia intracellularis</em></td>
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<tr>
<td>Identify laboratories and their respective testing capabilities prior to need. Some laboratories are able to offer a wide array of diagnostic tests by forwarding received samples to other laboratories. In time sensitive situations, diagnostic test results can be expedited by submitting samples directly to the laboratory that will be performing the test.</td>
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<tr>
<td>The laboratory should be accredited by the <a href="https://www.aavld.org">American Association of Veterinary Laboratory Diagnosticians (AAVLD)</a>. List of <a href="https://www.aavld.org">Accredited Laboratories</a>,</td>
</tr>
</tbody>
</table>

| **Sampling** |
| Contact the laboratory for information on preferred sample handling for specific tests. |
| Submit fecal and blood samples cooled, but not frozen, in leak-proof containers with icepacks. |
| Anaerobic transport media (Port-A-Cul™) should not be refrigerated, but shipped at room temperature. Do not allow to freeze. |

| **Sample Collection** |
| **Feces** |
| Collect feces into urine cup or another suitable leak proof container. Ensure no leakage of sample possible during storage and transportation (cross-contamination) |
| Sample Volume: |
| 3–10 grams of feces or 3–10 cc of liquid feces (diarrhea) |
| 2–3 rectal swabs if a quantity of feces is not obtainable |
| Swabs for PCR synthetic swab (no cotton tipped swabs) snapped off into a red top tube (RTT). Avoid swabs with wooden applicators. Swabs from culturettes are acceptable, but avoid putting swab back into media/gel. No culture media, viral transport media, or saline solution is necessary. |
| Swabs for aerobic culture should be placed in aerobic bacterial transport media (Amies Transport Media, Para-Pak®) |
| Swabs for anaerobic culture should be placed in anaerobic transport media (Port-A-Cul™). (Note: Port-A-Cul™ supports both aerobic and anaerobic culture) |

| **Blood** |
| Submission of whole blood (EDTA) and serum. Avoid freezing of whole blood. |

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Contact the laboratory for information on the preferred shipping protocol for certain types of specimens, hours of operation for receiving shipments, and whether a laboratory is open over weekends or on holidays for receipt of diagnostic materials.

- Use the appropriate submission form provided by the laboratory (FAX or internet download)
- All samples for viral isolation must be shipped cold (in an insulated container with cold packs), and preferably arrive within 24 hours of dispatch. Always use overnight or same-day delivery services
- Frozen samples must be shipped on dry ice or several frozen freezer packs and appropriate packaging. (Check with commercial shipping company for specific shipping requirements; noncompliance can result in the package being rejected.)
- If possible, do not ship on Fridays; not every lab is open to receive samples on weekends. Refrigerate or freeze samples and ship on the following Monday
- Virus containing samples are considered hazardous and must comply with IATA guidelines for air shipping or Postal Service guidelines (see below)
- For local or in-state laboratories, a courier service may be more expedient and less complicated than using a commercial shipping company. (Notify lab if courier service is being used and determine specifically where and to whom the sample is to be delivered.)

**Information on sample transport**

**Shipping non-hazardous agents**

**Safe shipping of samples**

**For shipping by air, call** FedEx or alternative company Dangerous Goods/Hazardous Materials Hotline. The number to call in the case of FedEx is 1-800-463-3339 (press 81) for further information.

The **United States Postal Service** has set specific guidelines for the proper packaging of biological materials for shipment. Diagnostic specimens, potentially infectious specimens, and other animal products are considered hazardous materials. Shipping services may refuse to handle any package that shows signs of internal breakage, spillage, or dampness. The sender could be held legally responsible for improperly packaged specimens; careful packaging is essential.

**Shipping guidelines**

- Submit all specimens in a leak proof container
- Enclose completed submission forms in a separate plastic bag and place between the inner sample container and the outer shipping container

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- Surround that container with sufficient absorbent material to absorb any possible leakage
- Containers must then be enclosed in a sturdy and sealed secondary container (cardboard, plastic, styrofoam, etc.)
- If more than one primary container is placed in the secondary packaging, each container must be wrapped with enough absorbent material to ensure that contact is prevented and that the absorbent material can absorb the entire contents of all materials being shipped
- Fresh tissue samples should be placed in individual, well-sealed, heavy plastic bags or other containers. Double bag to prevent leakage
- Ship refrigerated and frozen specimens with adequate cold packs to ensure samples are kept cool or frozen during shipment

**Do not**
- submit samples in syringes
- include needles in samples submitted
- use ice cubes or water filled plastic bags as refrigerant
- wrap submission form(s) around sample(s)

**Diagnosis**
Proceed based on disease-specific information:
- *Salmonella* spp.
- *Clostridial enteritis* – *Clostridium difficile, Clostridium perfringens*
- *Potomac Horse Fever* – *Neorickettsia risticii*
- Rotavirus (RTA)
- *Lawsonia intracellularis*
- Coronavirus (ECoV)

**No Diagnosis**
- Maintain biosecurity measures for 21–28 days after onset of last clinical case
- Consult infectious disease expert