Tick Borne Diseases of the Horse

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**Disclosures**

Employed by Boehringer Ingelheim Animal Health

**Lyme Disease in Horses**

- Fast growing vector-borne infectious disease in the U.S.
- 25X increase in annual reported cases since 1982 surveillance
- ~30,000 cases per year reported to CDC
  - Only a fraction reported
- CDC studies
  - Large Commercial Laboratories in U.S. (2008)
    - 288,000 (240,000-444,000)
  - Incidence of Clinician Diagnosed Lyme (2005-2010)
    - 320,000 (296,000-376,000)
  - Suggest ~300,000 cases diagnosed per year

**CDC Facts on Lyme in Humans**

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CDC Statistics

- 95% of confirmed cases were reported from 14 states (2015)

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Lyme Disease History

- First described in 1975 in 51 human patients Lyme, Old Lyme and East Haddam, CT
- Multiple joint arthritis
- 1982 Willy Burgdorfer PhD student identified organism – Borrelia burgdorferi
- Found within deer tick (Ixodes scapularis) on Long Island, NY
- Slender, spirally undulating bacteria (spirochete).
- 1984 first reported lyme arthritis in dog

Life cycle

- Ixodes scapularis/Ixodes pacificus (deer tick/black legged tick)
  - Requires 3 different hosts to complete life cycle (egg to adult)
  - Each feeding stage requires blood meal
  - Adult female lays the egg→larva
  - Larval stage→nymph
  - Nymph stage→adult
  - Each stage is capable of causing infection
- B. burgdorferi in hindgut
- Must be attached 24-48 hrs.
- B. burgdorferi, B. mayonii, B. miyamotoi
Clinical observations/concerns

• Controversial opinions exist about occurrence in horses
• Often non-specific, vague clinical signs
• High seroprevalence in clinically healthy horses
• Inducing disease experimentally?
• Accumulating evidence supports Lyme diagnosis in horses

Clinical signs

• Behavioral and lameness
  – Shifting/multiple limb lameness
• Poor performance
• Swollen joints/Arthritis
• Hyperesthesia
• Uveitis
• Pseudolymphoma
• Neuroborreliosis
  – Gait abnormalities, ataxia, depression, head tilt, CN deficits, neck stiffness, encephalitis
  – Lymphocytic pleocytosis in CSF and Ab’s to B. burgdorferi

Diagnostics

• Potential for infection
  – Horse lives or was in endemic area
  – Clinical signs of disease
  – Absence of other disease
  – Positive screening/confirmatory test
• ELISA/IFA
• Western blot
• C6 ELISA
• Multiplex

Diagnostics

• ELISA or IFA- IgG measurement
• High titer
  – Exposed
  – Infected and sick
  – Recently cleared infection
• Low titer
  – Not infected
  – Cleared infection
  – Hasn’t mounted immune response yet
• Cross reactivity with other spirochetal organisms
• Titers do not equate to clinical signs or degree

Diagnostics

• Western blot
  – Designed as confirmatory test for IFA; more specific
  – Detects many antigenic patterns of B. burgdorferi
  – No cross reactivity
  – Indicate common vaccine responses
  – Serial samples useful in following infection
  – Serum, CSF, synovial fluid
  – Limitations:
    • Subjective
    • Reference laboratory only
    • Expensive
• C6 ELISA: 3Dx, 4Dx, Quantitative C6
  • C6 (IR6) Invariable region of the VslE protein
  • C6 gene expressed in mammalian tissues
  • Specific for infection (live organism)
  • Avoids vaccination confusion
  • Consistent with WB 3wks post infection (100%)
  • High agreement between Ab’s of C6 and OspF as markers for infection
  • Effective monitoring infection
    • Ab decrease indicates reduction of parasite load/viability

Diagnostics

- Multiplex assay
  - Cornell AHDC 2011
  - Fluorescent bead technology
  - High sensitivity and specificity
  - Limited labor/quick turn around

Treatment

- Doxycycline
  - 10mg/kg bwt q12hrs. per os
  - G. I. disturbances in rare cases
- Minoxycline
  - 4mg/kg bwt q12hrs. per os
- Oxytetracycline
  - 6.6-11mg/kg bwt q24hrs i.v.
  - Dehydration/pre-existing renal dysfunction
- Ceftiofur
  - 2-4mg/kg bwt q12hrs i.v. or i.m.

Prevention

- Tick control
  - Limit possible exposure
  - Vegetation control
  - Careful examination and removal of ticks
  - Acaricides (dog wipes)
  - Limit mouse population
  - float with pyrethrin

- Vaccination
  - No current vaccine available for the horse
  - Dog vaccine has been used

Diagnostics

- Multiplex assay
- Osp A: vaccination? Can be transiently positive w/nat. infect.
- Osp C: early infection (~3wks.); decline by 7-11wks., undetectable by 4-5mos.
- Osp F: late/chronic infection (5-8wks.); remains elevated.
  Osp F+ Osp C- infected for at least 5mos.
- Assessment of treatment-6wks. post

Diagnostics

- PCR
  - B. burgdorferi rarely present in blood
  - Useful in testing
    - Urt, synovial fluid, CSF
    - Synovial tissue biopsies
    - Renal/other biopsies
    - Necropsy specimens

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### Canine vaccines

- 2 Killed, adjuvanted, whole cell, bivalent bacterins (Osp A & C)
  - Duramune® Lyme (Elanco)
  - Novibac® Lyme (Merck)
- 1 Recombinant, non-adjuvanted, plasma derived, sub-unit vaccine (Osp A)
  - Recombitek® Lyme (BI)

### Field samples from vaccinated horses (n=65)
- Evaluated Osp A, C, and F immune responses
- Similar Osp A responses
- <65% had MFI >2,000 1-3 mos. post vaccination
- Virtually gone by 7-9 mos.

### Three vaccine experiments

#### Experiment #1
- Responses to 3 canine vaccines
  - Naïve Icelandic horses
  - Vaccinated Days 0, 25, and 108
  - Osp A values low for all vaccines; <2000 MFI by 16 weeks
  - Osp C elevated with 1 killed, whole cell bacterin

#### Experiment #2
- Vaccine dose
  - Previously non-vaccinated
  - Whole cell (Osp A/C): 2ml vs. 1ml
  - Osp A and C responses: p<0.01 and p<0.0001
  - Rapid decline regardless of dose

#### Experiment #3
- Response of prevaccinated horses
  - 2 doses of Recombitek
  - Earlier onset of Osp A and increased Ab longevity (>18 weeks)
  - By 24 weeks all <2,000 MFI
• Experiment #4
  – Route of vaccination
    • SC or IM route
    • Short lived Osp A response (16wks.) regardless of route of administration

• Take home’s:
  • Vaccination has potential to protect horses from infection
  • Osp A Ab’s correlate with protection in many species (Fikrig E, Barthold SW, Kantor FS, Flavell RA. J Infect Dis. 1991 Dec; 164(6):1224-7.)
  • Maybe important tool in endemic locations
  • Transient and inconsistent results in horses
  • Increasing dose and booster frequency may be beneficial
    • Lyme nephritis

Equine Granulocytic Anaplasmosis

• Anaplasma phagocytophilum
• Gram negative bacterium
  – Formerly known as Ehrlichia equi.
  – Tick borne (Ixodes spp.)
• Infectious, non-contagious, seasonal disease
  – Originally observed in no. California
  – Recognized in many states
  – Seen in Europe, Africa, and South America
• Effects wide range of hosts
  – Horses, burros, llamas, rodents
• Human granulocytic anaplasmosis (HGE)
  – Similar strain as horses

Clinical signs

• Fever
• Anorexia, depression, reluctance to move
• Limb edema
• Icterus, petechiation
• Ataxia
• Clinical signs more severe in adults than in horses <4 y.o.
  – Younger horses <1 often only experience fever

Anaplasma phagocytophilum

• Clinicopathologic changes
  – Leukopenia (neutropenia)
  – Thrombocytopenia
    • Mechanism?
  – Hyperfibrinogenemia
  – Mild anemia
  – Intra-cytoplasmic granular inclusion bodies (morulae)
    • Cocobacillary organisms within membrane bound vacuoles
    • Present in neutrophils and eosinophils
    • Giemsa or Wrights stain
Transmission

- Infection through exposure to Ixodes sp
  - Ixodes scapularis and Ixodes pacificus
- Can be transmitted experimentally
  - Whole blood from infected horses/people with HGA
- Incubation period is 1-3 weeks
- Zoonotic risk of infection to people via horses has not been observed; although infection in both appear to be with strains of the same agent

Diagnosis

- Demonstration of cytoplasmic inclusion bodies
  - Few first 48hrs.
  - Increase to 30-40% neutrophils by 3-5 days
- PCR on whole blood
- Paired serology
- SNAP 4Dx
- Differential diagnoses include:
  - Viral encephalitis
  - Primary liver disease
  - EIA
  - Purpura hemorrhagica
  - EVA

Treatment

- Oxytetracycline 7 mg/kg q24 for 8 days
- Penicillin, chloramphenicol, streptomycin no inhibitory effect
- Banamine
- Short term corticosteroid treatment severely ataxic
  - Dexamethasone, 20mg/day, for 2-3 days
- Supportive wraps/stall rest
- Shorter duration treatment may see relapse
- Resolution without therapy
- Solid immunity (>2yrs.) upon recovery

Control

- Tick control measures are mandatory for control of disease.
- No vaccine currently available

Equine Piroplasmosis

- Obligate intra-erythrocytic protozoan parasites
  - Theileria equi (formerly Babesia equi)
  - Babesia caballi
- Can also affect donkeys, mules, zebras
  - Clinical disease rare
- Reportable disease
Equine Piroplasmosis

• Transmission
  – Ticks and biting insects
  – Iatrogenic

• Incubation period
  – 10-30 days B. caballi
  – 12-19 days T. equi

• Persistently infected, inapparent carriers
  – Sequestration of organism and immune evasion
    – Capillaries, CNS vasculature, bone marrow?
    – T. equi infection lifelong?
    – B. caballi may persist for years/lifetime
      – Accounts of self clearance of organism

• Life cycle of B. caballi and T. equi

Equine Piroplasmosis

• Ticks are the definitive hosts and vector
  – Must undergo sexual development within the tick

• Relatively few species of ticks can support

• Competent tick vectors in the U.S. include:
  ➢ Theileria equi
    • Amblyomma mixtum (Cayenne tick, formerly known as A. cajennese)
    • Dermacentor variabilis (American dog tick)
  ➢ Babesia caballi
    • Dermacentor nitens (Tropical horse tick)
    • Dermacentor albipictus (Winter tick)

Epidemiology

• Widespread in tropical and subtropical areas
  – Endemic in Africa, Central and South America, Caribbean, Middle East, Asia, Mediterranean

• T. equi tends to be higher prevalence
  – No cross immunity
  – Mixed infections may occur

• First documented U.S. case in FL. 1961
• Increased surveillance led to disease free status 1988
• Several outbreaks have put status in jeopardy over the years

Clinical signs

• Clinical disease can manifest in different forms

• Acute signs can be non-specific
  – High fevers (104°F)
  – Lethargy, anorexia, weight loss
  – Peripheral edema
  – Petechiations due to thrombocytopenia
  – Hemolytic anemia
    • Fever, tachycardia, tachypnea, weakness, pigmenturia (hemoglobinuria/bilirubinuria)
  – GI complications (colic, impactions, diarrhea)
  – Pneumonia, pulmonary edema, cardiac arrhythmias
  – CNS signs: ataxia, myalgia, seizures
  – Laminitis

• Fulminant (peracute)
  – Collapse and sudden death with overwhelming T. equi infections
    • Intro of naïve horses into endemic area of France resulted in 99% fatality rate
  – Neonatal foals infected in-utero can exhibit clinical signs at birth or 2-3 days of age
    • Weakness, decreased suckling, progress to adult signs
  – B. caballi-foal and neonatal infections reported but rare

• Chronic infection non-specific signs

• Commonly inapparent carriers
  – Reservoirs for transmission
    • Biggest concern for non-endemic countries

Clinical signs
Diagnosis

- Hemolytic anemia, thrombocytopenia, leukocytosis
- Giemsa stained blood smear
- Serology
  - Competitive ELISA: official U.S. test
  - Indirect fluorescent antibody: also accepted by OIE
- PCR
  - Nested and real-time
  - Research only

Treatment

- Imidocarb dipropionate
  - Mode of action uncertain
  - 4.4 mg/kg IM q72hrs for 4 treatments will clear B. caballi
  - T. equi more refractory to treatment
  - Dose dependent hepatotoxicity and nephrotoxicity
  - Colic, diarrhea, sweating
  - Buscopan, glycopyrolate, atropine prevent
- Oxytetracycline
  - 4-6 mg/kg IV for 7 days
  - Effective against T. equi but not B. caballi
- Ponazuril inhibits T. equi in vitro
  - No in-vivo work to date
- Fluids, NSAID's, blood transfusions

Prevention

- Impossible in endemic countries
- Non-endemic countries- regulation of equine movement
- OIE regulatory efforts successful
  - Isolated cases continue to occur in non-endemic locations
  - Rarely associated with tick transmission
  - Iatrogenic—blood contaminated equipment and practices involving needle sharing, blood doping/ transfusions, improperly sterilized surgical, dental, and tattoo equipment

Seroprevalence of Piro in the U.S.
USDA-APHIS 2009

- 15,300 samples were tested
  - 35 EIA sample labs in 34 states submitted samples
- Seroprevalence for B. caballi was 54 horses per 100,000 (0.054%)
- Seroprevalence for T. equi was 7 horses per 100,000 (0.007%)

Piroplasmosis in Florida-2008

- 7 year y.o. QH presented to UF
  - Lethargy, edema, icterus
  - Theileria equi identified
- Subsequent investigation of 210 horses on 25 premises
  - 20 T. equi positives found on 7 premises
  - 7 had clinical signs consistent with EP
  - No B. caballi positives
- Dermacentor variabilis
  - All ticks negative on testing
- Suggestive of iatrogenic spread of T. equi
  - Contaminated needles and blood doping
EP in Texas - 2009

- South TX. Ranch
- Mare presented with clinical signs consistent with EP
  - Positive on cELISA for T. equi
- 292/360 positive on index premises
- Trace out found 117 positives in 15 states
- Tick transmission
  - Competent ticks present:
    - Amblyomma mixtum
    - Dermacentor variabilis
- Many states test horses from TX

Options for EP Positive Horses

- Permanent quarantine
- Euthanasia
- Export from country
- Long term quarantine with enrollment in USDA treatment research program (introduced Feb. 2013)
  - Dr. Don Knowles, WSU

USDA Treatment and Research Program

- Introduced in 2013
- Gives T. equi positive horses a chance for release from quarantine
  - Must complete treatment protocol
  - Must be shown to be clear of organism by series of methods over time
  - Test negative on all diagnostics
- Of 262 horses that have tested positive since 2009
  - 162 died or euthanized
  - 18 exported
  - 55 enrolled in USDA program
  - 26 have been released
- Texas outbreak
  - 163 enrolled
  - 140 have met all negative test requirements