UNIVERSITY OF GEORGIA

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College of Veterinary Medicine
Department of Pathology

Case number: C25-007376

Presenter: Daria Mrugala, VMD (University of Georgia, College of Veterinary Medicine, Department of Pathology, 501 D. W. Brooks Drive, Athens, GA 30602, daria.mrugala@uga.edu)

Contributors & Acknowledgements: Samantha Schlemmer, DVM, MS, DACVP; original diagnosticians & clinicians: Satoshi Suzuki, BVSc, MS, PhD, DJCVP, Jennifer Steinberg, DVM, DACVP, Chelsea Folmar, DVM, & Rachel Pfeifle, DVM, MS, DACVIM (LAIM)

Presenter title: Diagnostic Laboratory Medicine Intern

Signalment: 6-month-old quarter horse colt weanling

History: The colt was presented to UGA after multiple days of diarrhea and evaluation by referring veterinarians. Prior treatments included flunixin meglumine (intermittently administered following pyrexia, 104 °F), fenbendazole (after suspected ascarids were seen in the diarrhea), and gentamicin and trimethoprim/sulfadiazine (after neutropenia was identified on blood work).

Clinical findings: Physical exam in-hospital revealed rectal temperature of 101.8 °F, tachycardia (72 beats per minute), tachypnea (36 breaths per minute), mildly hyperemic and dry mucous membranes with a prolonged capillary refill time of 2.5 seconds, prolonged skin turgor, and delayed jugular refill. Wheezes and crackles were audible across all lung fields, and mild bilateral mucoid nasal discharge was appreciated. Borborygmi were increased and fluid-sounding in all abdominal quadrants. Thoracic ultrasound revealed diffuse comet tails and multifocal areas of consolidation bilaterally. Abdominal ultrasound showed thickening of the large colon wall and increased intraluminal fluid in the large and small intestines. Complete blood cell count abnormalities included leukopenia 3.9 $x10^3$ cells/uL (RI: 5.1-10.5) characterized by a neutropenia 0.66 $x10^3$ cells/uL (RI: 2.71-6.25) and increased band neutrophils 0.03 (RI: none), and hyperfibrinogenemia 600 mg/dL (RI: 100-400). Blood gas and biochemistry abnormalities included hyponatremia 120 mmol/L (RI: 132.3-138.9), hypochloremia 96.5 mmol/L (RI: 99.9-108.3), hypocalcemia (ionized) 1.36 mmol/L (RI: 1.38-1.67), hypomagnesemia (ionized) 0.45 mmol/L (RI: 0.47-0.72), hyperlactatemia 4.6 mmol/L (RI: 0.5-1.5), azotemia (BUN 29 mg/dL [RI: 11-22], creatinine 2.6 mg/dL [RI: 0.8-1.7]), panhypoproteinemia 3.7 g/dL (RI: 5.8-7.8; hypoalbuminemia 1.3 g/dL [RI: 2.5-3.3], hypoglobulinemia 2.4 g/dL [RI: 2.8-5.1]), increased AST activity 399 U/L (RI: 193-375), and hypertriglyceridemia 165 mg/dL (RI: 16-61).

A transtracheal wash cytology sample is provided for your evaluation.

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Transtracheal wash (TTW) cytologic description: The sample is moderately to highly cellular on a thick, proteinaceous background and contains numerous fungal hyphae, mixed population bacteria, and few *Alternaria* and nematode larvae, along with large numbers of degenerated and necrotic inflammatory cells including neutrophils, macrophages, and lymphocytes. The hyphae measure 2.5 µm wide by 30-40 µm long with thin cellular walls, septations, acute angle branching, and brush border-like fruiting bodies. Some of them appear necrotic or degenerate. Bacteria are a mixed population of rods, cocci, and coccobacilli and are present extracellularly and within inflammatory cells. Larvae visualized are amphophilic and slender with a pointed head and tail measuring approximately 150-300 µm long and approximately 25 µm wide. No overt neoplastic cells are found.

TTW opinion: Marked mixed inflammation with fungal and bacterial infection and intralesional larvae and evidence of necrosis

Ancillary testing: Multiple organisms were present on aerobic culture of TTW fluid including moderate growth of *Chryseobacterium gleum* and moderate growth of *Streptococcus ovis* which is usually considered a nonpathogenic commensal. Anaerobic culture of TTW fluid was impeded by moderate to heavy filamentous fungal overgrowth. Fungal culture was not pursued. The animal declined and euthanasia was performed, but postmortem examination was not elected.

Comment: In a study analyzing TTW and bronchoalveolar lavages in horses, the most common isolated fungi were *Penicillium*, *Aspergillus*, *Rhizomucor*, and *Candida*.[1] These can be environmental contaminants or opportunists and contributory factors of fungal infection include stabling of horses in moist conditions, providing dusty hay, prolonged administration of antimicrobials that upset the microfloral balance, and existence of an immunosuppressive state due to severe malnutrition, congenital immunodeficiency, acquired immunodeficiency, endocrinopathy, neoplasia, or long-term administration of immunosuppressive drugs.[2]

The most common gram-negative bacterial species isolated following airway fluid sampling are *Escherichia coli*, *Klebsiella*, *Enterobacter*, *Pasteurella*, *Actinobacillus*, and *Bordetella*. Common gram-positive bacteria are *Streptococcus zooepidemicus*, *Streptococcus pneumoniae*, and *Rhodococcus equi*.[3] The most

common anaerobes are *Bacillus* (facultative anaerobe), *Peptostreptococcus*, and *Fusobacterium*.[3] *S. zooepidemicus*, beta-hemolytic *Streptococcus* spp, *S. pneumoniae*, *Actinobacillus*, *Bordetella,* and *Pasteurella* isolates are considered to primary pathogens,[3] while *Pseudomonas* are typically opportunistic an may also reflect contamination from poor sampling technique.[4] *Chrysobacterium gleum* is a gram-negative aerobic bacterium and is an uncommon isolate but in humans is considered a significant opportunistic pathogen when identified.[5] *Streptococcus ovis* is primarily recognized as a commensal organism in horses but there is a report of abortion in a mare housed with sheep where *S. ovis* was isolated from the lung of the aborted fetus.[6]

The most common parasites affecting the lungs of horses are *Dictyocaulus arnfieldi* in mature horses and *Parascaris equorum* in foals and weanlings.[7] *D. arnfieldi* is considered a true lungworm, with donkeys serving as the natural reservoir host; ingested infective larvae migrate through the gastrointestinal tract to the lungs where they mature, are coughed up, swallowed and passed in the feces. *D. arnfieldi* eggs are 90 µm in diameter, first stage larvae are 450 µm in length and can grow up to 7 cm as adults. Egg, larval, and adult stages can be found in the lungs.[7] In contrast, *P. equorum* is an intestinal roundworm whose larvae undergo hepatic-pulmonary migration after ingestion, passing through the liver and lungs, swallowed, returning to the intestine to complete maturation. *P. equorum* eggs encapsulate the L3 larval stages and are 100 µm in diameter.[7,8] Larval stages are found in the lungs and mature adults that molt in the intestines can reach up to 2 feet long.[8] Fecal analysis can facilitate parasite diagnosis and often requires complementary methods (e.g., flotation for ova and Baermann for larvae); however, false negatives may occur in pre-patent infections, so results should be interpreted with other clinical and diagnostic findings (e.g., airway wash/lavage cytology, airway and intestinal endoscopy). Unfortunately, fecal analysis was not performed in the present case, but the history and signalment are most suggestive of *P. equorum* infection.

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