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Pneumonia Caused by *Klebsiella* spp. in 46 Horses

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Background: *Klebsiella* spp. are implicated as a common cause of bacterial pneumonia in horses, but few reports describe clinical presentation and disease progression.

Hypothesis/Objectives: To describe the signalment, clinicopathologic data, radiographic and ultrasonographic findings, antimicrobial susceptibility, outcome, and pathologic lesions associated with *Klebsiella* spp. pneumonia in horses.

Animals: Forty-six horses from which *Klebsiella* spp. was isolated from the lower respiratory tract.

Methods: Retrospective study. Medical records from 1993 to 2013 at the William R. Pritchard Veterinary Medical Teaching Hospital, University of California, Davis were reviewed. Exact logistic regression was performed to determine if any variables were associated with survival to hospital discharge.

Results: Survival in horses <1 year old was 73%. Overall survival in adults was 63%. For adults in which *Klebsiella pneumoniae* was the primary isolate, survival was 52%. Mechanical ventilation preceded development of pneumonia in 11 horses. Complications occurred in 25/46 horses, with thrombophlebitis and laminitis occurring most frequently. Multi-drug resistance was found in 47% of bacterial isolates. Variables that significantly impacted survival included hemorrhagic nasal discharge, laminitis, and thoracic radiographs with a sharp demarcation between marked caudal pulmonary alveolar infiltration and more normal-appearing caudodorsal lung.

Conclusions and Clinical Importance: *Klebsiella* spp. should be considered as a differential diagnosis for horses presenting with hemorrhagic pneumonia and for horses developing pneumonia after mechanical ventilation. Multi-drug resistance is common. Prognosis for survival generally is fair, but is guarded for adult horses in which *K. pneumoniae* is isolated as the primary organism.

Key words: Hemorrhagic pneumonia; Multi-drug resistance; Respiratory infection.

Klebsiella spp. are commonly implicated as a cause of bacterial pneumonia in horses, but few reports describe the clinical presentation and progression of disease attributable to this organism.[1–5] Although single case reports discuss the clinical presentation of pneumonia caused by *Escherichia coli* or *Actinobacillus* spp.,[6,7] to the authors' knowledge no large retrospective studies describe the clinical course of pneumonia in which the primary bacterial isolate is *Klebsiella* spp.

Klebsiella spp. are gram-negative, rod-shaped, facultative anaerobic bacteria. The organism is ubiquitous in the environment and is part of the normal urogenital and intestinal microflora of the horse.[5,8] In human medicine, *Klebsiella* spp. are a common cause of nosocomial pneumonia in patients who have received mechanical ventilation.[9,10] Pneumonia is an occasional complication of mechanical ventilation in the horse, but an association with a particular bacterial species has not been reported. Gram-positive bacteria are present in most cases of pneumonia in horses, and frequently are accompanied by gram-negative and anaerobic bacteria.[11–13] Although previous studies have shown that isolation of anaerobic bacteria and *E. coli* negatively impact survival in horses with bacterial pneumonia,[11,14] no other bacterial species has been associated with poor prognosis.[11,14]

Comprehensive reports of pneumonia in horses caused by *Klebsiella* spp. are lacking in the veterinary literature. The first objective of our study was to describe the signalment, clinical signs, clinicopathologic data, diagnostic imaging findings, and antimicrobial susceptibility patterns of horses with *Klebsiella* spp. pneumonia and to determine if any of these variables are associated with survival to hospital discharge. The second objective of this study was to describe gross pathologic and histopathologic lesions of horses that died or were euthanized as a result of *Klebsiella* spp. pneumonia.

Abbreviations:

aPTT	activated partial thromboplastin time
CI	confidence interval
FDPs	fibrin degradation products
MDR	multi-drug resistant
OR	odds ratio
SIRS	systemic inflammatory response syndrome
WBC	white blood cell

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Materials and Methods

Animals

In this retrospective study, medical records from the University of California, Davis William R. Pritchard Veterinary Medical Teaching Hospital between 1993 and 2013 were reviewed. Horses were included in the study population if *Klebsiella* spp. was isolated by bacterial culture as a primary or secondary organism from the lower respiratory tract. In all cases, bacterial culture was performed on samples obtained by percutaneous transtracheal wash, thoracocentesis, or lung tissue collected postmortem. For aerobic cultures, samples were inoculated on 5% defibrinated sheep blood, chocolate, and MacConkey agars and incubated at 35°C in 5% CO_2. For anaerobic cultures, samples were inoculated on Brucella blood agar and incubated under anaerobic conditions. Bacterial identification was accomplished by conventional biochemical reactions using tubed media and spot testing. An organism was considered the primary if it was the sole genus detected or was highest in number in a mixed bacterial infection. Antimicrobial susceptibility testing and interpretations were performed using the broth microdilution method[a] according to the methodology described by the Clinical Laboratories Standards Institute.[15] Antimicrobial susceptibility results were examined, and an isolate was determined to be multidrug resistant (MDR) if it was resistant to drugs in ≥3 antimicrobial classes (eg, beta lactams, aminoglycosides, tetracyclines, fluoroquinolones, potentiated sulfonamides).

Data Collection

Data including signalment, history of a predisposing event (eg, transport, mechanical ventilation, strenuous exercise), presenting complaint, physical examination findings, radiographic and ultrasonographic findings, complications, and outcome were compiled. Adult horses were considered febrile if rectal temperature was elevated ≥38.6 C, and a cut-off temperature of ≥38.9 C was used for foals. Horses were classified as tachycardic if heart rate exceeded 48 bpm for adults or 100 bpm for foals and were considered tachypneic if respiratory rate was >30 breaths/min for adults or >60 breaths/min for foals.

When available, data including CBC, serum biochemical analysis, and coagulation panel results were reviewed. In horses that did not survive, gross pathologic and histopathologic findings were evaluated.

Diagnostic imaging records, including thoracic ultrasound and radiographs, were reviewed. Because of variable reporting of ultrasonographic findings and quality of archived ultrasound images, results were reported as the presence or absence of pleural effusion. Thoracic radiographs obtained during a patient's initial period of hospitalization were reviewed by a board-certified radiologist (AY). Digital radiographic images obtained after 2004 were reviewed using Digital Imaging and Communications in Medicine (DICOM) eFILM viewing software,[b] whereas film images obtained before 2004 were viewed using a standard lightbox and hot light. A radiographic study was considered complete if most or all of the pulmonary parenchyma could be visualized even if a specific projection was absent from the study. A complete study usually required 2–4 radiographic projections, depending on the age and size of the patient. Most studies were unilateral, although some had a combination of both left and right lateral projections. Single projections or incomplete studies were not evaluated when a complete study from the contralateral side was available. The pulmonary parenchyma was divided into quadrants before evaluation: Quadrant 1—dorsal to the silhouettes of the heart base and junction of the brachycephalic trunk with the right atrium, cranial to the carina; Quadrant 2—dorsal to the silhouettes of the heart base and dorsal margin of the caudal vena cava, caudal to the carina; Quadrant 3—ventral to the silhouettes of the heart base and junction of the brachycephalic trunk with the right atrium, cranial to the carina and, Quadrant 4—ventral to the silhouettes of the heart base and dorsal margin of the caudal vena cava, caudal to the carina. Each quadrant was assessed for the presence of abnormal pulmonary pattern(s) (bronchial, interstitial, or alveolar) and severity of the pattern(s). If complete thoracic studies with both right and left lateral projections were available, the highest score was recorded for each pattern. The pulmonary parenchyma also was assessed for the presence of masses, bullae, or emphysema. Pleural involvement was recorded if the pleural margin appeared thickened or if there was an observable fluid line. Thoracic radiographs also were evaluated for the presence of visible hilar lymph nodes as well as tracheal or cardiovascular abnormalities.

Data Analysis

Exact logistic regression was performed to determine each variable's impact on survival to discharge. Variables analyzed included age, physical examination, history of predisposing factors, *Klebsiella* species isolated, presence of multi-drug resistance, mixed bacterial or anaerobic infection, clinicopathologic data (total white blood cell [WBC] count, presence of immature neutrophils, thrombocytopenia, hyperfibrinogenemia), and development of complications. Results are reported as odds ratios (OR) and 95% confidence intervals (CI). A variable was considered to be significantly associated with survival to discharge if the OR was >1 with a corresponding P value of ≤ .05.

Results

Case Histories and Physical Examination Findings

Forty-six horses met the criteria for inclusion. Horses represented 8 breeds, which included Thoroughbred (16), Quarter Horse (15), Arabian (5), Paint (3), Morgan (2), Standardbred (2), Draft or Draft cross (2), and Appaloosa (1). This breed distribution was similar to that of our hospital population. Ages ranged from 12 hours to 24 years. Horses were divided into 2 groups based on age. Nineteen horses were <1 year of age (median, 2.9 months; range, 12 hours to 8 months) and were classified as foals, 27 were adults (median, 6.5 years; range, 1.5–24.6 years).

Data including age, presenting complaint, presence of hemorrhagic nasal discharge, disease or treatment complications, *Klebsiella* spp. isolated, mechanical ventilation, and survival are outlined in Table S1. Fever was found in 10/23 adults for which temperature was recorded (range, 37.2–40.4 C) and 11/19 foals (range, 95–39.2 C). Tachycardia was present in 13/20 adults and 7/19 foals. Tachypnea was found in 10/20 adults and 6/19 foals. Hemorrhagic nasal discharge was present in 12/27 adults and 3/19 foals. Mechanical ventilation during inhalation anesthesia to facilitate surgical procedures preceded the development of pneumonia in 11 horses (8 adults, 3 foals). Recent strenuous exercise was reported in 9 adult horses, and recent transport was reported in 7 adults.

Clinicopathologic Findings

Complete blood count was performed in 25/27 adults and 19/19 foals. Hyperfibrinogenemia >400 mg/dL was

found in 17/25 adults (range, 200–1,000 mg/dL) and 12/19 foals (range, 300–1,000 mg/dL). Total WBC count in adults ranged from 3,900 to 42,900/μL (normal reference: 5,000–11,600/μL) and was abnormal in 18/25 adults. Leukocytosis was present in 17 adults and characterized by neutrophilia. One adult was leukopenic and neutropenic. Band neutrophils were found in 11 adult horses, and myelocytes were found in 4 adult horses with 1 adult also having metamyelocytes. Total WBC count in foals ranged from 670 to 43,840/μL (reference range, 5,300–14,000/μL) and was abnormal in 6/19 foals. Leukocytosis was found in 3 foals and characterized by neutrophilia. Leukopenia characterized by neutropenia was found in 3 foals. Band neutrophils were present in 6 foals, and metamyelocytes were present in 2. Thrombocytopenia (platelets $< 100{,}000 \times 10^3$/μL, no evidence of platelet aggregation) was found in 4 adults and 3 foals. Evaluation of coagulation parameters was performed in 6 horses, and all were abnormal. Prothrombin time, activated partial thromboplastin time (aPTT), and fibrin degradation products (FDPs) were measured in 5 horses. Prothrombin time was prolonged in 3 horses, aPTT was prolonged in 1 horse, and FDPs were increased in 1 horse. Antithrombin III concentrations were tested in 1 horse and were low. Serum biochemical analysis was performed in 38/46 horses and did not show any consistent abnormalities.

Diagnostic Imaging

Results of ultrasonographic examination were recorded in 18 horses (10 adults, 8 foals). Pleural effusion was identified using ultrasound in 5/10 adults and 7/8 foals, and by thoracic radiography in 4/15 adults. All horses with radiographic evidence of pleural effusion also had pleural effusion that was visualized ultrasonographically. One horse had pleural effusion that was found using ultrasound, but was not identified on radiographs, likely because of thoracocentesis and drainage of pleural effusion performed before radiography.

Thoracic radiographs were available for review for 27/46 horses (15/27 adults, 12/19 foals). No specific common radiographic pattern could be identified; most radiographs indicated a combination of bronchial, interstitial, and alveolar patterns of variable severity. For the 15 adult horses with thoracic radiographs, the most common location of radiographic pulmonary pathology was in the caudodorsal (13/13) quadrant, with interstitial reticular (12/13) and alveolar (6/13) patterns occurring most frequently. Five adult horses had an abrupt line of demarcation in the caudodorsal lung fields with a short zone of transition between the most severely affected caudal pulmonary parenchyma and more normal appearing pulmonary opacity of the caudodorsal lung tip (Fig 1). The caudoventral lung was radiographically abnormal in 8/12 adult horses, with an alveolar pattern predominating (7/8). The cranioventral (3/11) and craniodorsal (2/13) quadrants were less frequently affected. Soft tissue opacities with a gas-fluid interface that were suspicious of pulmonary abscesses were identified in 2 adults.

Of the 12 foals with radiographs, 11 were abnormal. The caudodorsal (11/11) and caudoventral (9/11) lung fields were most consistently abnormal. A reticular interstitial pattern was the most common abnormality found, but bronchial, alveolar, and nodular interstitial patterns also were seen. One foal had evidence of pulmonary abscessation caudoventrally. The craniodorsal (7/11) and cranioventral (5/11) lung fields were less frequently affected. Two foals had evidence of hilar lymph node enlargement.

Microbiological Examination

Klebsiella pneumoniae was isolated in 40 horses; *Klebsiella oxytoca* was isolated in 6 horses. *Klebsiella pneumoniae* was identified as the primary organism in 35/40 horses: 21 were adults and 14 were foals (Table S1). Antimicrobial susceptibility testing was performed for 41 isolates (Table 1). Multi-drug resistance was found in 47% (19/41) of isolates with 17 of the MDR isolates identified as *K. pneumoniae*. Antimicrobial resistance was found most frequently to tetracycline (52.3%), gentamicin (48.8%), trimethoprim sulfamethoxazole (41.5%), and chloramphenicol (39.5%). Mixed bacterial infections occurred in 31 horses. In addition to *Klebsiella* spp., organisms isolated in adults included *Strep-*

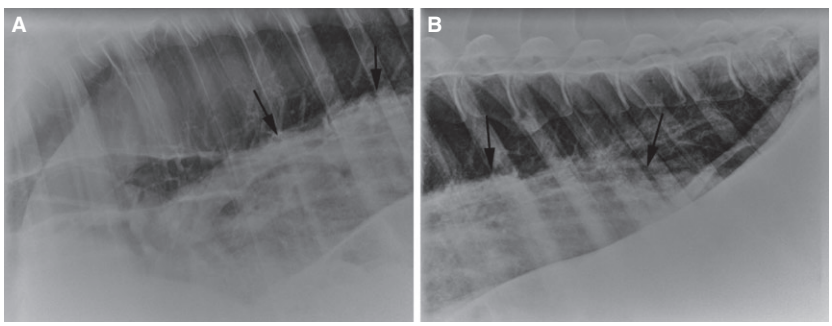

Fig 1. Lateral radiographic images of the dorsal (**A**) and caudodorsal (**B**) thorax showing a sharply demarcated region of increased soft tissue opacity within the dorsal and caudal lungs. The most caudodorsal region of the affected lung remains air filled. The increased soft tissue opacity correlated with pulmonary infarction and edema; a line of demarcation was appreciated at the time of postmortem.

tococcus equi ssp. *zooepidemicus* (7), *E. coli* (3), *Actinobacillus* spp. (3), *Pseudomonas aeruginosa* (2), *Pasteurella* spp. (2), *Staphylococcus aureus* (2), *Enterobacter* spp., *Prevotella* spp., *Streptococcus* spp., and *Acinetobacter baumannii*. Anaerobic bacteria isolated from adults included *Clostridium* spp. (4), *Fusobacterium* spp. (2), *Porphyromonos macacae*, and *Peptostreptococcus* spp. Bacterial organisms cultured in foals included *E. coli* (3), *Enterococcus casseliflavus, S. aureus, Rhodococcus equi, Enterobacter cloacae, Proteus* spp., and *Burkholderia cepacia*. Anaerobic bacteria isolated from foals included *Clostridium* spp., and *Prevotella* spp.

Outcome

Overall survival to discharge was 70% (32/46); 4 horses died and 10 were euthanized while in the hospital. Survival in horses <1 year of age was 79% (15/19). Overall survival to discharge in adults was 63% (17/27). For adults in which *K. pneumoniae* was isolated as the primary organism, survival was 52% (11/21). Although age appeared to be a factor in survival, the difference in survival between adults and foals was not statistically different. Complications associated with pneumonia or antimicrobial treatment occurred in 25/46 horses, with thrombophlebitis (10/25) and laminitis (7/25) occurring most frequently (Table S1).

Results of exact logistic regression are outlined in Table 2. Factors significantly associated with nonsurvival to discharge included presence of hemorrhagic nasal discharge (OR = 5.01, 95% CI = 1.01–29.75; P = .049), development of any complication associated with disease or antimicrobial treatment (OR = 6.03, 95% CI = 1.05–65.3, P = .043), laminitis (OR = 16.6,, 95% CI = 1.56–886, P = .013), and radiographic evidence of a sharp line of demarcation in the caudodorsal lung field with severe alveolar infiltrate ventrally (OR = 21.6, 95% CI = 2.5–∞, P = .004). Tachycardia (OR = 1.63, 95% CI = 0.814–352, P = .09), coinfection with anaerobic bacteria (OR = 5.14, 95% CI = 0.818–39.9, P = .088), and leukopenia (OR = 10.5, 95% CI = 0.903–571, P = .065) were associated with increased risk of death, but not significantly. The remainder of the reported variables as well as severity and distribution of radiographic abnormalities were not significantly associated with survival.

Postmortem examination and histopathology

Of the 14 horses that died or were euthanized as a result of *Klebsiella* spp. pneumonia, 12 horses (8 adults, 4 foals) underwent postmortem examination. All 4 horses with radiographs that disclosed a sharp line of demarcation with severe ventral pulmonary consolidation had a corresponding line of demarcation grossly, and discrete, dark red regions consistent with infarction (Fig 2A). Six horses had serosanguineous or yellow-green, cloudy pleural effusion. Pleural thickening, roughening, fibrin tags, or some combination of these were described in 5 horses. A gross description of lung lesions was not provided for 1 horse.

Microscopically, all horses had fibrinosuppurative broncho- or bronchointerstitial pneumonia affecting much of the ventral aspect of the lung, and 8 horses had histologic evidence of pleuritis, 2 of which were chronic (Fig 2B). There was microscopic thrombosis of pulmonary vessels (Fig 2C), pulmonary infarction or some combination of these in 8 horses. Of the 8 horses with pulmonary thrombosis or infarction, 6 were adults and 1 was a neonate. Five horses had discrete pulmonary abscesses, and 1 additional horse had abun-

Table 1. Antimicrobial susceptibility data for *Klebsiella* spp. obtained from the lower airway.

Antimicrobial	Number Tested	MIC Range, μg/mL	MIC50,[a] μg/mL	MIC90,[a] μg/mL	% Susceptible[b]
Amikacin	41	≤0.5 to >32	1	8	97.6
Amoxicillin/clavulanic acid	24	≤2 to >32	8	>16	50
Ampicillin	41	4 to >32	>16	>32	2.4
Cefazolin	17	≤2 to >16	≤2	>16	70.6
Cefotaxime	5	≤0.5 to <4	N/A[c]	N/A	100
Cefpodoxime	5	≤2 to >16	N/A	N/A	80
Cefoxitin	2	≤0.5 to >16	N/A	N/A	50
Cephalothin	26	≤2 to >32	16	>32	46.2
Cefixime	1	≤0.5	N/A	N/A	100
Ceftizoxime	34	≤0.5 to 8	≤0.5	4	100
Ceftiofur	38	≤0.25 to 8	0.5	4	81.6
Chloramphenicol	38	≤4 to >32	4	>32	60.5
Doxycycline	4	≤1 to 2	N/A	N/A	100
Enrofloxacin	38	≤0.25 to 8	≤0.25	≤0.5	97.4
Gentamicin	41	≤0.25 to >16	≤1	>16	51.2
Tetracycline	36	≤1 to >16	8	>16	47.2
Imipenem	6	≤0.25 to ≤1	N/A	N/A	100
Ticarcillin/clavulanic acid	41	≤4 to >128	≤16	128	73.2
Trimethoprim/sulfa	41	≤0.25 to >4	0.5	>4	58.5

[a]MIC50 and MIC90 represent the concentration of antimicrobial at which 50 or 90% of isolates were inhibited respectively.
[b]Intermediately susceptible and resistant isolates were excluded.
[c]Not applicable as MIC values for <10 isolates were available.

Table 2. Prognostic significance of history, physical examination, microbiological, and clinicopathologic results, presence of complications, and diagnostic imaging findings.

Variable	Survival Odds Ratio	95% Confidence Interval	*P*-Value
Predisposing factors			
Mechanical ventilation	0.821	0.118–4.35	1.0
Strenuous exercise	0.269	0.005–2.52	.417
Transport	1.63	0.200–11.7	.858
Physical examination			
Tachycardia	7.18	0.814–352	.09
Fever	0.544	0.097–2.61	.6
Hemorrhagic discharge*	5.01	1.01–29.8	.049
Microbiological results			
Klebsiella pneumoniae	2.31	0.383–25.3	.537
Klebsiella oxytoca	0.931	0–12.3	.959
Multi-drug resistance	0.908	0.189–4.62	1.0
Mixed bacterial infection	0.979	0.206–5.40	1.0
Anaerobic infection	5.14	0.818–39.9	.088
Clinicopathologic results			
Fibrinogen > 400	1.04	0.235–5.02	1.0
Leukocytosis	0.638	0.147–2.74	.698
Leukopenia	10.5	0.903–571	.065
Immature neutrophils	3.76	0.811–20.9	.102
Thrombocytopenia	0.804	0.068–5.6	1.0
Complications*	6.03	1.05–65.3	.043
Laminitis*	16.6	1.56–886	.013
Thrombophlebitis	0.432	0.039–2.66	.543
Colitis	1.62	0.12–16.3	.964
Diagnostic imaging			
Sharp line of demarcation*	21.6	2.5–∞	.004
Pleural effusion	1.96	0.385–10.3	.543
Pulmonary abscess	4.28	0.193–286	.535
Hilar lymph node enlargement	0.755	0.0–10.2	.837

*indicates statistical significance.

dant, but encapsulated intra-alveolar neutrophils. Bacteria were seen on hematoxylin and eosin-stained sections in 8 horses, and were confirmed as gram-negative rods in 4 cases by Brown and Brenn staining, Brown and Hopps staining or both. A mixed population (including gram-positive organisms) was demonstrated in 1 horse. Subjectively, organisms were better demonstrated with the Brown and Hopps method (Fig 2D) than the Brown and Brenn method.

Evidence of systemic inflammatory response syndrome (SIRS) characterized grossly by acute laminitis, multisystemic serosal hemorrhage, adrenal cortical hemorrhage, multicavitary effusion (peritoneal, pericardial), or some combination of these, was evident in 4 adults and 3 foals. Eleven of 12 horses that underwent postmortem examination had evidence of additional organs affected by *K. pneumoniae* or septicemic sequelae, with lesions including peritonitis, pericarditis, hepatic and splenic necrosis, diffuse lymphadenopathy (hyperplasia or lymphoid depletion), embolic suppurative nephritis, diffuse thrombosis, and laminitis. The horse without systemic sequelae had multisystemic T-cell lymphoma, which was considered to be unrelated to pulmonary disease.

Discussion

Survival to discharge in horses with *Klebsiella* spp. pneumonia was 70%. Previous studies report survival of horses with bacterial pneumonia as low as 46%[11] with more current studies reporting survival in up to 90% of cases.[11,16–18] In our study, survival to discharge in horses <1 year of age was 79% (15/19), whereas overall survival to discharge in adults was 63% (17/27). Although age group was not significantly associated with survival, prognosis for adults infected with *K. pneumoniae* as the primary organism was poor, with survival in this population decreased at 52% (11/21). In contrast, 80% (12/15) of foals with *K. pneumoniae* survived to discharge. Previous studies have identified laminitis and diarrhea as the most common nonrespiratory causes of euthanasia in horses with pneumonia.[11] In the present study, the odds of nonsurvival in horses developing any complication associated with pneumonia or antimicrobial treatment were 6 times higher than in horses that did not develop complications. The odds of death or euthanasia in horses that developed laminitis were 16 times higher than those that did not. Because laminitis rarely occurs in horses <1 year of age, the decreased survival in the adult population may have been a consequence of laminitis, which could have contributed to the decision to euthanize. The presence of hemorrhagic nasal discharge also was associated with nonsurvival, and was found in 12/27 adults as compared to 3/19 foals. Hemorrhagic nasal discharge likely was caused by severe pulmonary pathology and may have been an indication of more severe disease in adults than foals, which resulted in lower survival in adults. Additionally, our study identified that radiographs that showed a sharp line of demarcation with severe ventral pulmonary consolidation were associated with a 100% case fatality rate and corresponded to the presence of infarcted lung on postmortem examination. Because this radiographic pattern was not found in any foals in this study population, it may indicate either less severe disease in foals as compared to adults, or a difference in immune response between the 2 populations that resulted in dissimilarity in disease outcome.

Klebsiella spp. infection should be considered in horses presenting with signs of hemorrhagic pneumonia, because 15/46 horses presented with hemorrhagic nasal discharge. Although nasal discharge may have represented epistaxis because of coagulopathy, discharge often was reported to be dark in nature and to be increased after coughing, indicating that it may have originated from the lower respiratory tract. The clinical presentation of *K. pneumoniae* in humans is similar to that observed in the horses included in this study and includes a sudden onset of high fever, SIRS, and hemoptysis ("currant jelly sputum").[19] Causes of hemorrhagic nasal discharge in horses include severe pulmonary disease that compromises the pulmonary

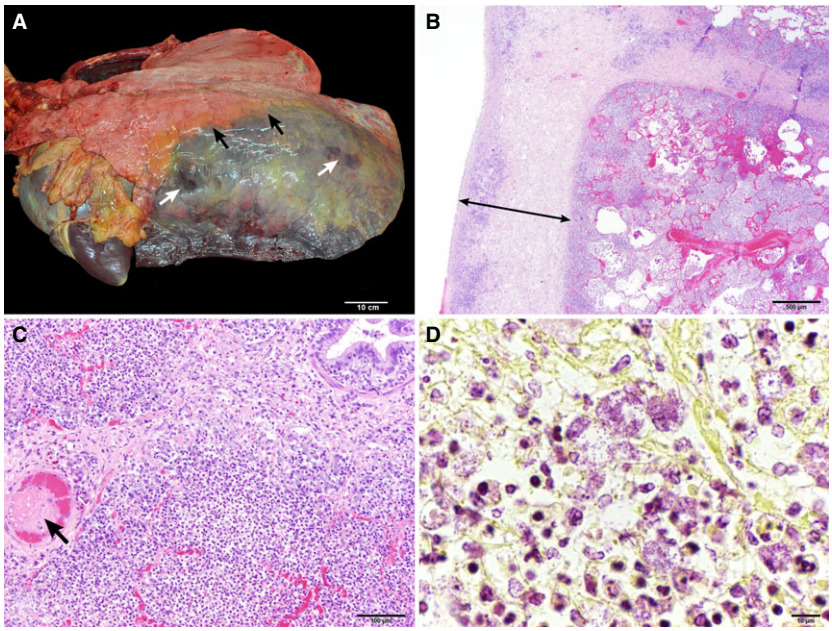

Fig 2. (**A**) Gross photograph of the lungs from a horse with *Klebsiella pneumoniae* pneumonia. Note the sharp line of demarcation (black arrows) between consolidated and inflamed ventral regions and more normal dorsal regions. Yellow discoloration of the pleura is fibrin, and there are foci of pulmonary hemorrhage (white arrows). Cranial is to the left. (**B**) Photomicrograph of affected lung. The pleural surface is reactive, and there is marked expansion of the subpleura by edema and fibrosis (double sided arrow). To the right of the figure is peripheral lung that is effaced by suppurative inflammation. 4×, H&E. (**C**) Photomicrograph of affected lung, with a small vessel fibrin thrombus (arrow), severe suppurative pneumonia. 10×, H&E. (**D**) Tissue Gram stain highlighting short, 1–2 μm, gram-negative (magenta) rods, both intra- and extracellularly. Some appear more coccoid, because of end-on orientation. 60×, Brown and Hopps method.

vasculature, pulmonary infarction, systemic coagulopathy, as well as upper airway disease. Six horses were tested for coagulopathy, and all were abnormal, but not all horses with hemorrhagic nasal discharge were tested. The pathophysiology of lung injury caused by *Klebsiella* spp. is not known, but endotoxin might increase pulmonary vascular permeability and result in acute lung injury and subsequent hemorrhagic pneumonia.[9,20] In our study, the presence of hemorrhagic nasal discharge was a negative prognostic indicator, with horses that presented with discharge having odds of not surviving 5 times higher than horses without discharge. Previously reported causes of hemorrhagic pneumonia include *Actinobacillus* spp.,[6] *E. coli,*[7] and pulmonary infarction,[21] although in the case report describing *E. coli* pneumonia, *K. pneumoniae* also was isolated and hemorrhagic nasal discharge was not described.[6,7,21] In our study, 3 horses were coinfected with *Actinobacillus* spp. and none were reported to have hemorrhagic nasal discharge during hospitalization. In a retrospective study of acute pulmonary infarction in horses, *Klebsiella* spp. was found in the lower respiratory tract of 2/14 horses, but it was uncertain whether or not bacterial infection was primary or secondary to an acute thromboembolic event.[21] The pathogenesis of pulmonary infarction in horses often is unknown, but it may be caused by systemic dysregulation of the coagulation cascade that results in a thromboembolus that lodges in the capillary bed of the pulmonary system, or could be caused by local infection and subsequent release of endotoxin. Pulmonary infarction may have resulted in the sharp line of demarcation, with severe pulmonary infiltrates found caudally in 5 horses. All 5 horses that displayed this radiographic pattern also had hemorrhagic nasal discharge and did not survive. Postmortem examination was performed in 4/5 of these horses, and evidence of pulmonary infarction was present in all of them.

Complications occurred in 54% of horses in our study. Thrombophlebitis was the most common complication, occurring in 10 horses. Thrombophlebitis is an occasional result of IV injection and indwelling catheterization.[22] However, in critically ill patients, endotoxemia and disseminated intravascular coagulation are potential causes of thrombophlebitis as a result of consumption of anticoagulant proteins and release of procoagulant factors. The frequency of thrombophlebitis in this study is similar to the frequency of thrombophlebitis reported in horses with colitis and gastrointestinal disease, indicating IV catheterization, fluid treatment, endotoxemia, and activation of the coagulation cascade likely are contributing factors.[23,24] Of the 46 horses in this study, 17 had at least 2 clinical signs of endotoxemia and SIRS (eg, fever, tachycardia, tachypnea, leukopenia). Tachycardia and leukopenia were negatively associated with survival in our study population, but this association was not significant. Previous studies in foals found that a diagnosis of SIRS was associated with pulmonary infiltrates within the caudodorsal lung.[17] Interestingly, the most common distribution of radiographic lesions in adults and foals in this study was in the caudodorsal lung field. This finding may indicate that SIRS resulted in vascular and

pulmonary damage in the caudodosal lung fields, which have increased perfusion when compared to the remainder of the lung.[25,26] Evidence of sepsis or SIRS was apparent in 11/12 horses that underwent postmortem examination.

Mechanical ventilation during inhalation anesthesia to facilitate surgical procedures preceded the development of pneumonia in 11 horses. The cases occurred in both isolated incidents and temporal clusters, and may have resulted from *Klebsiella* spp. colonization of a ventilator or other anesthetic equipment. Two cases occurred in horses that received mechanical ventilation between August and September 1998 and 4 cases occurred within 1 week in January 1999. Additionally, in 2 separate incidents, 2 herdmates presented with similar clinical signs and subsequently were diagnosed with *K. pneumoniae*. *Klebsiella* spp. is a common cause of ventilator-associated pneumonia in human hospitals, as well as a community-acquired pathogen, in people who suffer from alcoholism or are otherwise immunosuppressed.[10,19] Of the 46 horses that developed *Klebsiella* pneumonia, 26 had a known predisposing factor including mechanical ventilation, strenuous exercise, or a history of prolonged travel. Although immune status is likely a factor in the development of pneumonia in horses, because of the retrospective nature of our study, the presence of an incompetent immune system (eg, pituitary pars intermedia dysfunction, neonatal IgG concentrations) was not consistently evaluated and could not be implicated as a predisposing factor. However, given the behavior of *Klebsiella* spp. in human hospitals and in immunosuppressed humans and the findings reported in our study, *Klebsiella* spp. should be considered a potential nosocomial pathogen in hospitalized horses and a potentially contagious pathogen in herd situations. Of the antibiotics commonly used in horses, resistance occurred most frequently to gentamicin (48.8%), chloramphenicol (39.5%), tetracycline (52.3%), and trimethoprim sulfamethoxazole (41.5%). Multi-drug resistance was found in 47% (19/41) of isolates. Multi-drug resistant strains of *K. pneumoniae* are a common finding in human medicine, because *Klebsiella* spp. is exceptionally adept at acquiring plasmids containing multiple antibiotic resistant genes on transposable elements.[27] The presence of genes that confer antimicrobial resistance to aminoglycosides, chloramphenicol, tetracyclines, and sulfa drugs has been documented, as has horizontal transmission of MDR *Klebsiella* spp. in humans and animals.[27–29] In our study, susceptibility of isolates to ceftiofur and amikacin was better, with 81.6 and 97.6% being susceptible, respectively, although resistance of *K. pneumoniae* to 3rd generation cephalosporins is a common problem in human medicine.[29] Interestingly, 100% of isolates were reported as susceptible to doxycycline, but only 4 isolates were evaluated. The Clinical and Laboratory Standards Institute susceptible breakpoint for doxycycline ($\leq$4 μg/mL for Enterobacteriacae) is substantially higher than plasma concentrations achieved in horses after PO administration and treatment should be reserved for isolates with an MIC $\leq$0.25 μg/mL.[30] Although MDR did not significantly impact survival, horses that were coinfected with anaerobes were less likely to survive. This finding corroborates findings from previous studies that identified anaerobic infection as a negative prognostic indicator.[11,14]

Limitations of this study include its retrospective nature, which resulted in variation in reported clinical findings and occasionally missing data. Additionally, antimicrobial susceptibility testing changed over time and not all isolates were tested against the same antimicrobials. Both conventional and digital radiographs were evaluated and included in the study, although image quality varied between the 2 imaging methods. Radiographs may not have been taken at the same point in clinical disease, making comparison among horses difficult. In 1 foal, thoracic radiographs were normal and were potentially performed too early in the clinical course of disease to detect pulmonary changes. Although review of ultrasonographic findings may have added to the description of pulmonary pathology, results were not included in this study because of the poor quality of archived images and differences in reporting results.

Klebsiella spp. should be considered as a differential diagnosis for horses presenting with hemorrhagic pneumonia and for horses developing pneumonia after mechanical ventilation. Multi-drug resistance was common in our study, and appropriate biosecurity measures should be taken to prevent environmental contamination and nosocomial infection as occurs in human hospitals.[10] In this study population, all horses with radiographs that disclosed a sharp line of demarcation with severe ventral pulmonary consolidation did not survive and had evidence of pulmonary infarction on necropsy.

Footnotes

[a] Sensititre, Fisher Scientific, Oakwood Village, OH
[b] eFILM, Carlsbad, CA

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Conflict of Interest Declaration: Authors disclose no conflict of interest.

Off-label Antimicrobial Declaration: Authors declare no off-label use of antimicrobials.

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Supporting Information

Additional Supporting Information may be found online in Supporting Information:

Table S1. Signalment, presenting complaint, clinical signs, complications, microbiologic results, and survival of horses with *Klebsiella* spp. pneumonia.