

Acute Feedlot Arthritis Associated with Distinct Strains of *Mycoplasma bovis*

R. F. Rosenbusch
Veterinary Medical Research Institute
College of Veterinary Medicine
Iowa State University, Ames, IA

Abstract

Acute feedlot arthritis outbreaks have been reported recently throughout the Midwestern US. A common feature of these outbreaks has been poor responsiveness to antimicrobial therapy and the isolation of *Mycoplasma bovis* from joint fluid specimens. Morbidity and mortality of these outbreaks reached significant levels, and prompted an investigation of the mycoplasma strains recovered. Four strains of *M. bovis* recovered from acute arthritis cases were compared with 5 strains of *M. bovis* recovered from enzootic pneumonia cases. Invasive ability of these strains was compared *in vitro* using tracheal mucosa explants, and 1 strain of each type was inoculated intratracheally in calves. Arthritis-associated strains were more invasive *in vitro*. One arthritis-associated strain did not produce mycoplasmaemia in 4 infected calves, but was recovered from nasal mucosa. In contrast, a respiratory strain of *M. bovis* gave reproducible mycoplasmaemia but was not recovered from nasal mucosa. Phenotypic differences among strains of *M. bovis* warrant further study of the pathogenic and immunogenic properties of strains of *M. bovis*.

Acute mycoplasmal arthritis is reported in numerous species of animals and in man and can be caused by several species of mycoplasma. In every case, the mycoplasma involved is capable of invading mucosal surfaces and reaching joints after a brief mycoplasmaemia. This invasive capability is a poorly understood pathogenic feature of certain species of mycoplasmas, and it appears to be related to the ability of these mycoplasmas to traverse intercellular spaces. In cattle, mycoplasmal arthritis has been reported in association with mastitis or pneumonia caused by *mycoplasma bovis*.¹ A report of *M. bovis* arthritis in feedlot cattle in the absence of respiratory disease has been made.² Acute feedlot arthritis outbreaks have been reported throughout the US recently, and the common thread to these outbreaks is the poor responsiveness to antimicrobial therapy and the isolation of *M. bovis* from the joints of affected cattle. It was of interest to compare the invasive properties of *M. bovis* strains obtained from recent acute arthritis cases and *M. bovis* strains obtained from classical enzootic pneumonia cases.

Materials and Methods

Outbreaks studied

Samples of joint fluids from steers in 7 feedlot outbreaks of arthritis were cultured for mycoplasmas and for routine bacteriological examination. Data from clinical incidence was obtained from attending veterinarians.

Mycoplasma strains

Four strains of *M. bovis* were isolates from cases of acute feedlot arthritis. Isolates were obtained from joint fluids of early acute cases, subcultured in modified Friis medium and stored frozen as aliquots at low passage after one clone-purification. Five strains of *M. bovis* obtained from classical cases of enzootic pneumonia were likewise prepared and stored.

Invasion in calves

Calves were obtained from a closed Hereford-cross herd that had been shown to be free of *M. bovis* infection. They were weaned and placed in individual isolation rooms prior to use for infection trials. Calves were 8 weeks to 8 months old at the time of use. Two calves of 8 weeks of age and 2 of 6-8 months of age were infected by transtracheal inoculation with 10⁹ Colony-Forming Units (CFU) of strain M23 (a respiratory *M. bovis* strain) in 25 ml of saline. Four calves of similar ages were infected in the same way with 10⁹ CFU of strain M45 (an arthritis-associated strain of *M. bovis*). Blood and nasal secretion samplings were obtained from days 1 through 17 to detect *M. bovis*. Calves were euthanized at 17 days after infection and the respiratory tract examined for gross and histopathological lesions as well as persistence of the mycoplasma.

In vitro invasion assay

Tracheal ring explants were prepared from day-old dairy calves and cultured for 2 days in rolling tubes with medium containing 50 U/ml of bacitracin. Explant

cultures were inoculated with 10^7 CFU/ml of the various *M. bovis* strains tested. Explants were harvested at 2 and 6 hours after infection by fixation of the mucosa in periodate-lysine-paraformaldehyde. Paraffin sections were reacted with a monoclonal antibody specific for *M. bovis* followed by a peroxidase-labeled immunohistochemistry reagent to detect the extent of mucosal invasion observed with each *M. bovis* strain.

Results

Outbreaks

M. bovis was recovered from joint fluids of steers affected with acute arthritis. Isolations were successful from early cases or unmedicated cases. No other mycoplasma species were recovered and no significant bacterial species were found in these samples. Four of the cases occurred in backgrounding feedlots and 3 others in finishing lots. Prior respiratory disease outbreaks were recorded in 3 of the 7 cases. Morbidity reported was from less than 1% to 30% of steers in a pen or feedlot and mortality varied from none to 100% of affected animals. Mortality was significant if there was concomitant respiratory disease and in heavy steers. Poor response to antibiotic treatment was reported in all cases studied.

Infection in calves

Invasion was readily observed in calves inoculated with the respiratory M23 strain of *M. bovis*. Mycoplasmaemia was detected from 1 to 6 days after infection and was associated with febrile reactions. Recovery of *M. bovis* from nasal swabs was not successful. Calves infected with the arthritis-associated M45 strain did not have detectable mycoplasmaemia or febrile reactions. The mycoplasma was easily recovered from nasal swabs. Small focal areas of pneumonia were distributed through the lungs in calves infected with either strain of *M. bovis*. Histopathological evaluation of gross lesions revealed catarrhal bronchiolitis and focal interstitial pneumonia.

In vitro invasion assay

Invasion of tracheal explant mucosa was readily detectable at 6 h after infection. Arthritis-associated strains of *M. bovis* appeared to invade the mucosae of explants more readily than the respiratory strains.

Conclusions

Recent acute outbreaks of feedlot arthritis have been associated with isolation of *M. bovis* from joint fluids. These outbreaks have been variably associated with respiratory involvement and are clearly differentiable from clinical cases of *M. bovis*-associated enzootic pneumonia. While enzootic pneumonia is seen to occur primarily in dairy calves of 2 to 6 months of age, the recent outbreaks have involved beef cattle in feedlots. Poor response to antibiotic treatments was reported for all 7 cases studied. Using an *in vitro* assay for mucosal invasion, arthritis-associated strains were shown to be more invasive. Assays for invasion performed *in vivo* in calves with 2 strains did not parallel these findings, although differences in pathogenesis of infection could be recognized between these strains. Phenotypic differences among strains of mycoplasma causing arthritis have been reported in cases of mycoplasmal arthritis caused by *Mycoplasma putrefasciens* in dairy goats.³ *In vitro* correlates of these strain differences have been described. Further characterization of arthritis-associated *M. bovis* strains is needed to define differences between the arthritis-associated strains and pneumonia-associated strains. In particular, the potential for cross-protective immunity among these strains is to be considered.

References

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