

Transmission of *Mannheimia haemolytica* from Domestic Sheep (*Ovis aries*) to Bighorn Sheep (*Ovis canadensis*): Unequivocal Demonstration with Green Fluorescent Protein-tagged Organisms

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Abstract: Previous studies have demonstrated that bighorn sheep (BHS) die of pneumonia when commingled with domestic sheep (DS). However, these studies did not conclusively prove the transmission of pathogens from DS to BHS. The objective of this study was to determine unambiguously whether *Mannheimia haemolytica*, an important respiratory pathogen of BHS, is transmitted from DS to BHS when they commingle. *M. haemolytica* was obtained from the pharynx of four DS and tagged with a plasmid carrying the genes for green fluorescent protein (*gfp*) and beta-lactamase (*bla*). Four DS colonized with the tagged bacteria were kept 30 ft apart from four BHS for one month. No symptoms of respiratory disease were observed during this period. The DS and BHS were then allowed to have fence line contact for two months. During this period three BHS contracted the tagged bacteria from the DS. At the end of two months the animals were allowed to commingle. One BHS died on day two, two died on day five, and the fourth one was euthanized on day nine following commingling. Lungs from all four BHS showed gross- and histo-pathological lesions characteristic of *M. haemolytica* pneumonia. *M. haemolytica* isolated from all four BHS were confirmed to be the tagged bacteria from the DS by their growth in ampicillin-containing growth medium, PCR-amplification of genes encoding GFP and Bla, and immunofluorescent staining of GFP. These results unequivocally prove transmission of *M. haemolytica* from DS to BHS which results in pneumonia and death of BHS.

Biennial Symposium of the Northern Wild Sheep and Goat Council 17:58; 2010

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